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(54) Title: NOVEL PLANT GENES AND USES THEREOF
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(57) Abstract

Homologues of the Arabidopsis NIM1 gene, which is involved in the signal transduction cascade leading to systemic acquired resistance (SAR), are isolated from Nicotiana tabacum (tobacco), Lycopersicon esculentum (tomato), Brassica napus (oilseed rape), Arabidopsis thaliana, Beta vulgaris (sugarbeet), Helianthus annuus (sunflower), and Solanum tuberosum (potato). The invention further concerns transformation vectors and processes for expressing the NIM1 homologues in transgenic plants to increase SAR gene expression and enhance broad spectrum disease resistance.

(57) Abrégé

L'invention concerne des homologues du gène Arabidopsis NIM1, impliqué dans la cascade de transduction des signaux menant à la résistance systémique acquise (RSA), qui sont isolés à partir de Nicotiana tabacum (tabac), de Lycopersicon esculentum (tomate), de Brassica napus (colza oléagineux), d'Arabidopsis thaliana, de Beta vulgaris (betterave à sucre), d'Helianthus annuus (tournesol) et de Solanum tuberosum (pomme de terre). L'invention concerne également des vecteurs de transformation et des processus permettant d'exprimer les homologues de NIM1 dans des végétaux transgéniques afin d'accroître l'expression du gène RSA et d'élargir le large spectre de résistance aux maladies.

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(57) **Abstract**

Homologues of the *Arabidopsis NIM1* gene, which is involved in the signal transduction cascade leading to systemic acquired resistance (SAR), are isolated from *Nicotiana tabacum* (tobacco), *Lycopersicon esculentum* (tomato), *Brassica napus* (oilseed rape), *Arabidopsis thaliana*, *Beta vulgaris* (sugarbeet), *Helianthus annuus* (sunflower), and *Solanum tuberosum* (potato). The invention further concerns transformation vectors and processes for expressing the *NIM1* homologues in transgenic plants to increase SAR gene expression and enhance broad spectrum disease resistance.

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Description

NOVEL PLANT GENES AND USES THEREOF

The present invention relates to broad-spectrum disease resistance in plants, including the phenomenon of systemic acquired resistance (SAR). More particularly, the present invention relates to the identification, isolation and characterization of homologues of the *Arabidopsis NIM1* gene involved in the signal transduction cascade leading to systemic acquired resistance in plants.

Plants are constantly challenged by a wide variety of pathogenic organisms including viruses, bacteria, fungi, and nematodes. Crop plants are particularly vulnerable because they are usually grown as genetically-uniform monocultures; when disease strikes, losses can be severe. However, most plants have their own innate mechanisms of defense against pathogenic organisms. Natural variation for resistance to plant pathogens has been identified by plant breeders and pathologists and bred into many crop plants. These natural disease resistance genes often provide high levels of resistance to or immunity against pathogens.

Systemic acquired resistance (SAR) is one component of the complex system plants use to defend themselves from pathogens (Hunt and Ryals, 1996; Ryals *et al.*, 1996). *See also,* U.S. Patent No. 5,614,395. SAR is a particularly important aspect of plant-pathogen responses because it is a pathogen-inducible, systemic resistance against a broad spectrum of infectious agents, including viruses, bacteria, and fungi. When the SAR signal transduction pathway is blocked, plants become more susceptible to pathogens that normally cause disease, and they also become susceptible to some infectious agents that would not normally cause disease (Gaffney *et al.*, 1993; Delaney *et al.*, 1994; Delaney *et al.*, 1995; Delaney, 1997; Bi *et al.*, 1995; Mauch-Mani and Slusarenko, 1996). These observations indicate that the SAR signal transduction pathway is critical for maintaining plant health.

Conceptually, the SAR response can be divided into two phases. In the initiation phase, a pathogen infection is recognized, and a signal is released that travels through the phloem to distant tissues. This systemic signal is perceived by target cells, which react by expression of both SAR genes and disease resistance. The maintenance phase of SAR refers to the period of time, from weeks up to the entire life of the plant, during which the plant is in a quasi steady state, and disease resistance is maintained (Ryals *et al.*, 1996).

Salicylic acid (SA) accumulation appears to be required for SAR signal transduction. Plants that cannot accumulate SA due to treatment with specific inhibitors, epigenetic repression of phenylalanine ammonia-lyase, or transgenic expression of salicylate hydroxylase, which specifically degrades SA, also cannot induce either SAR gene expression or disease resistance (Gaffney *et al.*, 1993; Delaney *et al.*, 1994; Mauch-Mani and Slusarenko, 1996; Maher *et al.*, 1994; Pallas *et al.*, 1996). Although it has been suggested that SA might serve as the systemic signal, this is currently controversial and, to date, all that is known for certain is that if SA cannot accumulate, then SAR signal transduction is blocked (Pallas *et al.*, 1996; Shulaev *et al.*, 1995; Vernooij *et al.*, 1994).

Recently, *Arabidopsis* has emerged as a model system to study SAR (Uknes *et al.*, 1992; Uknes *et al.*, 1993; Cameron *et al.*, 1994; Mauch-Mani and Slusarenko, 1994; Dempsey and Klessig, 1995). It has been demonstrated that SAR can be activated in *Arabidopsis* by both pathogens and chemicals, such as SA, 2,6-dichloroisonicotinic acid (INA) and benzo(1,2,3)thiadiazole-7-carbothioic acid *S*-methyl ester (BTH) (Uknes *et al.*, 1992; Vernooij *et al.*, 1995; Lawton *et al.*, 1996). Following treatment with either INA or BTH or pathogen infection, at least three pathogenesis-related (PR) protein genes, namely, PR-1, PR-2, and PR-5 are coordinately induced concomitant with the onset of resistance (Uknes *et al.*, 1992, 1993). In tobacco, the best characterized species, treatment with a pathogen or an immunization compound induces the expression of at least nine sets of genes (Ward *et al.*, 1991). Transgenic disease-resistant plants have been created by transforming plants with various SAR genes (U.S. Patent No. 5,614,395).

A number of *Arabidopsis* mutants have been isolated that have modified SAR signal transduction (Delaney, 1997) The first of these mutants are the so-called *lsd* (lesions simulating disease) mutants and *acd2* (accelerated cell death) (Dietrich *et al.*, 1994; Greenberg *et al.*, 1994). These mutants all have some degree of spontaneous necrotic lesion formation on their leaves, elevated levels of SA, mRNA accumulation for the SAR genes, and significantly enhanced disease resistance. At least seven different *lsd* mutants have been isolated and characterized (Dietrich *et al.*, 1994; Weymann *et al.*, 1995). Another interesting class of mutants are *cim* (constitutive immunity) mutants (Lawton *et al.*, 1993). *See also,* U.S. Patent No. 5,792,904 and International PCT Application WO 94/16077. Like *lsd* mutants and *acd2*, *cim* mutants have elevated SA and SAR gene expression and resistance, but in contrast to *lsd* or *acd2,* do not display detectable lesions on their leaves. *cpr1* (constitutive expresser of PR genes) may be a type of *cim* mutant;

however, because the presence of microscopic lesions on the leaves of *cpr1* has not been ruled out, *cpr1* might be a type of *lsd* mutant (Bowling *et al.*, 1994).

Mutants have also been isolated that are blocked in SAR signaling. *ndr1* (non-race-specific disease resistance) is a mutant that allows growth of both *Pseudomonas syringae* containing various avirulence genes and also normally avirulent isolates of *Peronospora parasitica* (Century *et al.*, 1995). Apparently this mutant is blocked early in SAR signaling. *npr1* (nonexpresser of PR genes) is a mutant that cannot induce expression of the SAR signaling pathway following INA treatment (Cao *et al.*, 1994). *eds* (enhanced disease susceptibility) mutants have been isolated based on their ability to support bacterial infection following inoculation of a low bacterial concentration (Glazebrook *et al.*, 1996; Parker *et al.*, 1996). Certain *eds* mutants are phenotypically very similar to *npr1*, and, recently, *eds5* and *eds53* have been shown to be allelic to *npr1* (Glazebrook *et al.*, 1996). *nim1* (noninducible immunity) is a mutant that supports *P. parasitica* (i.e., causal agent of downy mildew disease) growth following INA treatment (Delaney *et al.*, 1995; U.S. Patent No. 5,792,904). Although *nim1* can accumulate SA following pathogen infection, it cannot induce SAR gene expression or disease resistance, suggesting that the mutation blocks the pathway downstream of SA. *nim1* is also impaired in its ability to respond to INA or BTH, suggesting that the block exists downstream of the action of these chemicals (Delaney *et al.*, 1995; Lawton *et al.*, 1996).

Allelic *Arabidopsis* genes have been isolated and characterized, mutants of which are responsible for the *nim1* and *npr1* phenotypes, respectively (Ryals *et al.*, 1997; Cao *et al.*, 1997). The wild-type *NIM1* gene product is involved in the signal transduction cascade leading to both SAR and gene-for-gene disease resistance in *Arabidopsis* (Ryals *et al.*, 1997). Ryals *et al.*, 1997 also report the isolation of five additional alleles of *nim1* that show a range of phenotypes from weakly impaired in chemically induced PR-1 gene expression and fungal resistance to very strongly blocked. Transformation of the wild-type *NPR1* gene into *npr1* mutants not only complemented the mutations, restoring the responsiveness of SAR induction with respect to PR-gene expression and disease resistance, but also rendered the transgenic plants more resistant to infection by *P. syringae* in the absence of SAR induction (Cao *et al.*, 1997). WO 98/06748 describes the isolation of *NPR1* from *Arabidopsis* and a homologue from *Nicotiana glutinosa*. *See also,* WO 97/49822, WO 98/26082, and WO 98/29537.

Despite much research and the use of sophisticated and intensive crop protection measures, including genetic transformation of plants, losses due to disease remain in the billions of dollars annually. Therefore, there is a continuing need to develop new crop protection measures based on the ever-increasing understanding of the genetic basis for disease resistance in plants. In particular, there is a need for the identification, isolation, and characterization of homologues of the *Arabidopsis NIM1* gene from additional species of plants.

In describing the present invention, the following terms will be employed, and are intended to be defined as indicated below.

Associated With / Operatively Linked: Refers to two DNA sequences that are related physically or functionally. For example, a promoter or regulatory DNA sequence is said to be "associated with" a DNA sequence that codes for an RNA or a protein if the two sequences are operatively linked, or situated such that the regulator DNA sequence will affect the expression level of the coding or structural DNA sequence.

Chimeric Gene: A recombinant DNA sequence in which a promoter or regulatory DNA sequence is operatively linked to, or associated with, a DNA sequence that codes for an mRNA or which is expressed as a protein, such that the regulator DNA sequence is able to regulate transcription or expression of the associated DNA sequence. The regulator DNA sequence of the chimeric gene is not normally operatively linked to the associated DNA sequence as found in nature.

Coding Sequence: a nucleic acid sequence that is transcribed into RNA such as mRNA, rRNA, tRNA, snRNA, sense RNA or antisense RNA. Preferably the RNA is then translated in an organism to produce a protein.

Complementary: refers to two nucleotide sequences that comprise antiparallel nucleotide sequences capable of pairing with one another upon formation of hydrogen bonds between the complementary base residues in the antiparallel nucleotide sequences.

Expression: refers to the transcription and/or translation of an endogenous gene or a transgene in plants. In the case of antisense constructs, for example, expression may refer to the transcription of the antisense DNA only.

Expression Cassette: A nucleic acid sequence capable of directing expression of a particular nucleotide sequence in an appropriate host cell, comprising a promoter operatively linked to the nucleotide sequence of interest which is operatively linked to termination signals. It also typically comprises sequences required for proper translation of

the nucleotide sequence. The expression cassette comprising the nucleotide sequence of interest may be chimeric, meaning that at least one of its components is heterologous with respect to at least one of its other components. The expression cassette may also be one which is naturally occurring but has been obtained in a recombinant form useful for heterologous expression. Typically, however, the expression cassette is heterologous with respect to the host, i.e., the particular nucleic acid sequence of the expression cassette does not occur naturally in the host cell and must have been introduced into the host cell or an ancestor of the host cell by a transformation event. The expression of the nucleotide sequence in the expression cassette may be under the control of a constitutive promoter or of an inducible promoter which initiates transcription only when the host cell is exposed to some particular external stimulus. In the case of a multicellular organism, such as a plant, the promoter can also be specific to a particular tissue, or organ, or stage of development.

Gene: A defined region that is located within a genome and that, besides the aforementioned coding nucleic acid sequence, comprises other, primarily regulatory, nucleic acid sequences responsible for the control of the expression, that is to say the transcription and translation, of the coding portion. A gene may also comprise other 5' and 3' untranslated sequences and termination sequences. Further elements that may be present are, for example, introns.

Heterologous DNA Sequence: The terms "heterologous DNA sequence", "exogenous DNA segment" or "heterologous nucleic acid," as used herein, each refer to a sequence that originates from a source foreign to the particular host cell or, if from the same source, is modified from its original form. Thus, a heterologous gene in a host cell includes a gene that is endogenous to the particular host cell but has been modified through, for example, the use of DNA shuffling. The terms also includes non-naturally occurring multiple copies of a naturally occurring DNA sequence. Thus, the terms refer to a DNA segment that is foreign or heterologous to the cell, or homologous to the cell but in a position within the host cell nucleic acid in which the element is not ordinarily found. Exogenous DNA segments are expressed to yield exogenous polypeptides.

Homologous DNA Sequence: A DNA sequence naturally associated with a host cell into which it is introduced.

Isocoding: A nucleic acid sequence is isocoding with a reference nucleic acid sequence when the nucleic acid sequence encodes a polypeptide having the same amino acid sequence as the polypeptide encoded by the reference nucleic acid sequence.

Isolated: In the context of the present invention, an isolated nucleic acid molecule or an isolated enzyme is a nucleic acid molecule or enzyme that, by the hand of man, exists apart from its native environment and is therefore not a product of nature. An isolated nucleic acid molecule or enzyme may exist in a purified form or may exist in a non-native environment such as, for example, a recombinant host cell.

Minimal Promoter: promoter elements, particularly a TATA element, that are inactive or that have greatly reduced promoter activity in the absence of upstream activation. In the presence of a suitable transcription factor, the minimal promoter functions to permit transcription.

Native: refers to a gene that is present in the genome of an untransformed cell.

Naturally occurring: the term "naturally occurring" is used to describe an object that can be found in nature as distinct from being artificially produced by man. For example, a protein or nucleotide sequence present in an organism (including a virus), which can be isolated from a source in nature and which has not been intentionally modified by man in the laboratory, is naturally occurring.

NIM1: Gene described in Ryals *et al.*, 1997, which is involved in the SAR signal transduction cascade.

NIM1: Protein encoded by the *NIM1* gene

Nucleic acid: the term "nucleic acid" refers to deoxyribonucleotides or ribonucleotides and polymers thereof in either single- or double-stranded form. Unless specifically limited, the term encompasses nucleic acids containing known analogues of natural nucleotides which have similar binding properties as the reference nucleic acid and are metabolized in a manner similar to naturally occurring nucleotides. Unless otherwise indicated, a particular nucleic acid sequence also implicitly encompasses conservatively modified variants thereof *(e.g.* degenerate codon substitutions) and complementary sequences and as well as the sequence explicitly indicated. Specifically, degenerate codon substitutions may be achieved by generating sequences in which the third position of one or more selected (or all) codons is substituted with mixed-base and/or deoxyinosine residues (Batzer *et al., Nucleic Acid Res.* 19: 5081 (1991); Ohtsuka *et al., J. Biol. Chem.* 260: 2605-2608 (1985); Rossolini *et al., Mol. Cell. Probes* 8: 91-98 (1994)). The terms "nucleic acid" or "nucleic acid sequence" may also be used interchangeably with gene, cDNA, and mRNA encoded by a gene. In the context of the present invention, the nucleic acid molecule is preferably a segment of DNA. Nucleotides are indicated by their bases by the following standard abbreviations: adenine (A), cytosine (C), thymine (T), and guanine (G).

ORF: Open Reading Frame.

Plant: Any whole plant.

Plant Cell: Structural and physiological unit of a plant, comprising a protoplast and a cell wall. The plant cell may be in form of an isolated single cell or a cultured cell, or as a part of higher organized unit such as, for example, a plant tissue, a plant organ, or a whole plant.

Plant Cell Culture: Cultures of plant units such as, for example, protoplasts, cell culture cells, cells in plant tissues, pollen, pollen tubes, ovules, embryo sacs, zygotes and embryos at various stages of development.

Plant Material: Refers to leaves, stems, roots, flowers or flower parts, fruits, pollen, egg cells, zygotes, seeds, cuttings, cell or tissue cultures, or any other part or product of a plant.

Plant Organ: A distinct and visibly structured and differentiated part of a plant such as a root, stem, leaf, flower bud, or embryo.

Plant tissue: A group of plant cells organized into a structural and functional unit. Any tissue of a plant *in planta* or in culture is included. This term includes, but is not limited to, whole plants, plant organs, plant seeds, tissue culture and any groups of plant cells organized into structural and/or functional units. The use of this term in conjunction with, or in the absence of, any specific type of plant tissue as listed above or otherwise embraced by this definition is not intended to be exclusive of any other type of plant tissue.

Promoter: An untranslated DNA sequence upstream of the coding region that contains the binding site for RNA polymerase II and initiates transcription of the DNA. The promoter region may also include other elements that act as regulators of gene expression.

Protoplast: An isolated plant cell without a cell wall or with only parts of the cell wall.

Purified: the term "purified," when applied to a nucleic acid or protein, denotes that the nucleic acid or protein is essentially free of other cellular components with which it is associated in the natural state. It is preferably in a homogeneous state although it can be in either a dry or aqueous solution. Purity and homogeneity are typically determined using analytical chemistry techniques such as polyacrylamide gel electrophoresis or high performance liquid chromatography. A protein which is the predominant species present in a preparation is substantially purified. The term "purified" denotes that a nucleic acid or protein gives rise to essentially one band in an electrophoretic gel. Particularly, it means that the nucleic acid or protein is at least about 50% pure, more preferably at least about 85% pure, and most preferably at least about 99% pure.

Recombinant DNA molecule: a combination of DNA molecules that are joined together using recombinant DNA technology

Regulatory Elements: Sequences involved in controlling the expression of a nucleotide sequence. Regulatory elements comprise a promoter operably linked to the nucleotide sequence of interest and termination signals. They also typically encompass sequences required for proper translation of the nucleotide sequence.

Selectable marker gene: a gene whose expression in a plant cell gives the cell a selective advantage. The selective advantage possessed by the cells transformed with the selectable marker gene may be due to their ability to grow in the presence of a negative selective agent, such as an antibiotic or a herbicide, compared to the growth of non-transformed cells. The selective advantage possessed by the transformed cells, compared to non-transformed cells, may also be due to their enhanced or novel capacity to utilize an added compound as a nutrient, growth factor or energy source. Selectable marker gene also refers to a gene or a combination of genes whose expression in a plant cell gives the cell both, a negative and a positive selective advantage.

Significant Increase: an increase in enzymatic activity that is larger than the margin of error inherent in the measurement technique, preferably an increase by about 2-fold or greater of the activity of the wild-type enzyme in the presence of the inhibitor, more preferably an increase by about 5-fold or greater, and most preferably an increase by about 10-fold or greater.

The terms "identical" or percent "identity" in the context of two or more nucleic acid or protein sequences, refer to two or more sequences or subsequences that are the same or have a specified percentage of amino acid residues or nucleotides that are the same, when compared and aligned for maximum correspondence, as measured using one of the following sequence comparison algorithms or by visual inspection.

Substantially identical: the phrase "substantially identical," in the context of two nucleic acid or protein sequences, refers to two or more sequences or subsequences that have at least 60%, preferably 80%, more preferably 90-95%, and most preferably at least 99% nucleotide or amino acid residue identity, when compared and aligned for maximum correspondence, as measured using one of the following sequence comparison algorithms or by visual inspection. Preferably, the substantial identity exists over a region of the sequences that is at least about 50 residues in length, more preferably over a region of at least about 100 residues, and most preferably the sequences are substantially identical over at least about 150 residues. In a most preferred embodiment, the sequences are

substantially identical over the entire length of the coding regions. Furthermore, substantially identical nucleic acid or protein sequences perform substantially the same function.

For sequence comparison, typically one sequence acts as a reference sequence to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are input into a computer, subsequence coordinates are designated if necessary, and sequence algorithm program parameters are designated. The sequence comparison algorithm then calculates the percent sequence identity for the test sequence(s) relative to the reference sequence, based on the designated program parameters.

Optimal alignment of sequences for comparison can be conducted, e.g., by the local homology algorithm of Smith & Waterman, *Adv. Appl. Math.* 2: 482 (1981), by the homology alignment algorithm of Needleman & Wunsch, *J. Mol. Biol.* 48: 443 (1970), by the search for similarity method of Pearson & Lipman, *Proc. Nat'l. Acad. Sci. USA* 85: 2444 (1988), by computerized implementations of these algorithms (GAP, BESTFIT, FASTA, and TFASTA in the Wisconsin Genetics Software Package, Genetics Computer Group, 575 Science Dr., Madison, WI), or by visual inspection (*see generally,* Ausubel *et al., infra).*

One example of an algorithm that is suitable for determining percent sequence identity and sequence similarity is the BLAST algorithm, which is described in Altschul *et al., J. Mol. Biol.* 215: 403-410 (1990). Software for performing BLAST analyses is publicly available through the National Center for Biotechnology Information (http://www.ncbi.nlm.nih.gov/). This algorithm involves first identifying high scoring sequence pairs (HSPs) by identifying short words of length W in the query sequence, which either match or satisfy some positive-valued threshold score T when aligned with a word of the same length in a database sequence. T is referred to as the neighborhood word score threshold (Altschul *et al.*, 1990). These initial neighborhood word hits act as seeds for initiating searches to find longer HSPs containing them. The word hits are then extended in both directions along each sequence for as far as the cumulative alignment score can be increased. Cumulative scores are calculated using, for nucleotide sequences, the parameters M (reward score for a pair of matching residues; always > 0) and N (penalty score for mismatching residues; always < 0). For amino acid sequences, a scoring matrix is used to calculate the cumulative score. Extension of the word hits in each direction are halted when the cumulative alignment score falls off by the quantity X from its maximum achieved value, the cumulative score goes to zero or below due to the accumulation of one or more negative-scoring

residue alignments, or the end of either sequence is reached. The BLAST algorithm parameters W, T, and X determine the sensitivity and speed of the alignment. The BLASTN program (for nucleotide sequences) uses as defaults a wordlength (W) of 11, an expectation (E) of 10, a cutoff of 100, M=5, N=-4, and a comparison of both strands. For amino acid sequences, the BLASTP program uses as defaults a wordlength (W) of 3, an expectation (E) of 10, and the BLOSUM62 scoring matrix *(see* Henikoff & Henikoff, *Proc. Natl. Acad. Sci. USA* 89: 10915 (1989)).

In addition to calculating percent sequence identity, the BLAST algorithm also performs a statistical analysis of the similarity between two sequences *(see, e.g.,* Karlin & Altschul, *Proc. Nat'l. Acad. Sci. USA* 90: 5873-5787 (1993)). One measure of similarity provided by the BLAST algorithm is the smallest sum probability (P(N)), which provides an indication of the probability by which a match between two nucleotide or amino acid sequences would occur by chance. For example, a test nucleic acid sequence is considered similar to a reference sequence if the smallest sum probability in a comparison of the test nucleic acid sequence to the reference nucleic acid sequence is less than about 0.1, more preferably less than about 0.01, and most preferably less than about 0.001.

Another indication that two nucleic acid sequences are substantially identical is that the two molecules hybridize to each other under stringent conditions. The phrase "hybridizing specifically to" refers to the binding, duplexing, or hybridizing of a molecule only to a particular nucleotide sequence under stringent conditions when that sequence is present in a complex mixture (*e.g.,* total cellular) DNA or RNA. "Bind(s) substantially" refers to complementary hybridization between a probe nucleic acid and a target nucleic acid and embraces minor mismatches that can be accommodated by reducing the stringency of the hybridization media to achieve the desired detection of the target nucleic acid sequence.

"Stringent hybridization conditions" and "stringent hybridization wash conditions" in the context of nucleic acid hybridization experiments such as Southern and Northern hybridizations are sequence dependent, and are different under different environmental parameters. Longer sequences hybridize specifically at higher temperatures. An extensive guide to the hybridization of nucleic acids is found in Tijssen (1993) *Laboratory Techniques in Biochemistry and Molecular Biology-Hybridization with Nucleic* Acid Probes part I chapter 2 "Overview of principles of hybridization and the strategy of nucleic acid probe assays" Elsevier, New York. Generally, highly stringent hybridization and wash conditions are selected to be about 5ºC lower than the thermal melting point (T_m) for the specific sequence

at a defined ionic strength and pH. Typically, under "stringent conditions" a probe will hybridize to its target subsequence, but to no other sequences.

The T_m is the temperature (under defined ionic strength and pH) at which 50% of the target sequence hybridizes to a perfectly matched probe. Very stringent conditions are selected to be equal to the T_m for a particular probe. An example of stringent hybridization conditions for hybridization of complementary nucleic acids which have more than 100 complementary residues on a filter in a Southern or northern blot is 50% formamide with 1 mg of heparin at 42ºC, with the hybridization being carried out overnight. An example of highly stringent wash conditions is 0.1 5M NaCl at 72ºC for about 15 minutes. An example of stringent wash conditions is a 0.2x SSC wash at 65ºC for 15 minutes (*see,* Sambrook, *infra*, for a description of SSC buffer). Often, a high stringency wash is preceded by a low stringency wash to remove background probe signal. An example medium stringency wash for a duplex of, e.g., more than 100 nucleotides, is 1x SSC at 45ºC for 15 minutes. An example low stringency wash for a duplex of, *e.g.,* more than 100 nucleotides, is 4-6x SSC at 40ºC for 15 minutes. For short probes (*e.g.,* about 10 to 50 nucleotides), stringent conditions typically involve salt concentrations of less than about 1.0M Na ion, typically about 0.01 to 1.0 M Na ion concentration (or other salts) at pH 7.0 to 8.3, and the temperature is typically at least about 30ºC. Stringent conditions can also be achieved with the addition of destabilizing agents such as formamide. In general, a signal to noise ratio of 2x (or higher) than that observed for an unrelated probe in the particular hybridization assay indicates detection of a specific hybridization. Nucleic acids that do not hybridize to each other under stringent conditions are still substantially identical if the proteins that they encode are substantially identical. This occurs, e.g., when a copy of a nucleic acid is created using the maximum codon degeneracy permitted by the genetic code.

The following are examples of sets of hybridization/wash conditions that may be used to clone homologous nucleotide sequences that are substantially identical to reference nucleotide sequences of the present invention: a reference nucleotide sequence preferably hybridizes to the reference nucleotide sequence in 7% sodium dodecyl sulfate (SDS), 0.5 M $NaPO_4$, 1 mM EDTA at 50°C with washing in 2X SSC, 0.1% SDS at 50°C, more desirably in 7% sodium dodecyl sulfate (SDS), 0.5 M $NaPO_4$, 1 mM EDTA at 50°C with washing in 1X SSC, 0.1% SDS at 50°C, more desirably still in 7% sodium dodecyl sulfate (SDS), 0.5 M $NaPO_4$, 1 mM EDTA at 50°C with washing in 0.5X SSC, 0.1% SDS at 50°C, preferably in 7% sodium dodecyl sulfate (SDS), 0.5 M $NaPO_4$, 1 mM EDTA at 50°C with washing in 0.1X

SSC, 0.1% SDS at 50°C, more preferably in 7% sodium dodecyl sulfate (SDS), 0.5 M $NaPO_4$, 1 mM EDTA at 50°C with washing in 0.1X SSC, 0.1% SDS at 65°C.

A further indication that two nucleic acid sequences or proteins are substantially identical is that the protein encoded by the first nucleic acid is immunologically cross reactive with, or specifically binds to, the protein encoded by the second nucleic acid. Thus, a protein is typically substantially identical to a second protein, for example, where the two proteins differ only by conservative substitutions.

The phrase "specifically (or selectively) binds to an antibody," or "specifically (or selectively) immunoreactive with," when referring to a protein or peptide, refers to a binding reaction which is determinative of the presence of the protein in the presence of a heterogeneous population of proteins and other biologics. Thus, under designated immunoassay conditions, the specified antibodies bind to a particular protein and do not bind in a significant amount to other proteins present in the sample. Specific binding to an antibody under such conditions may require an antibody that is selected for its specificity for a particular protein. For example, antibodies raised to the protein with the amino acid sequence encoded by any of the nucleic acid sequences of the invention can be selected to obtain antibodies specifically immunoreactive with that protein and not with other proteins except for polymorphic variants. A variety of immunoassay formats may be used to select antibodies specifically immunoreactive with a particular protein. For example, solid-phase ELISA immunoassays, Western blots, or immunohistochemistry are routinely used to select monoclonal antibodies specifically immunoreactive with a protein. See Harlow and Lane (1988) *Antibodies, A Laboratory Manual,* Cold Spring Harbor Publications, New York "Harlow and Lane"), for a description of immunoassay formats and conditions that can be used to determine specific immunoreactivity. Typically a specific or selective reaction will be at least twice background signal or noise and more typically more than 10 to 100 times background.

"Conservatively modified variations" of a particular nucleic acid sequence refers to those nucleic acid sequences that encode identical or essentially identical amino acid sequences, or where the nucleic acid sequence does not encode an amino acid sequence, to essentially identical sequences. Because of the degeneracy of the genetic code, a large number of functionally identical nucleic acids encode any given polypeptide. For instance the codons CGT, CGC, CGA, CGG, AGA, and AGG all encode the amino acid arginine. Thus, at every position where an arginine is specified by a codon, the codon can be altered to any of the corresponding codons described without altering the encoded protein. Such

nucleic acid variations are "silent variations" which are one species of "conservatively modified variations." Every nucleic acid sequence described herein which encodes a protein also describes every possible silent variation, except where otherwise noted. One of skill will recognize that each codon in a nucleic acid (except ATG, which is ordinarily the only codon for methionine) can be modified to yield a functionally identical molecule by standard techniques. Accordingly, each "silent variation" of a nucleic acid which encodes a protein is implicit in each described sequence.

Furthermore, one of skill will recognize that individual substitutions deletions or additions that alter, add or delete a single amino acid or a small percentage of amino acids (typically less than 5%, more typically less than 1%) in an encoded sequence are "conservatively modified variations," where the alterations result in the substitution of an amino acid with a chemically similar amino acid. Conservative substitution tables providing functionally similar amino acids are well known in the art. The following five groups each contain amino acids that are conservative substitutions for one another: Aliphatic: Glycine (G), Alanine (A), Valine (V), Leucine (L), Isoleucine (I); Aromatic: Phenylalanine (F), Tyrosine (Y), Tryptophan (W); Sulfur-containing: Methionine (M), Cysteine (C); Basic: Arginine (R), Lysine (K), Histidine (H); Acidic: Aspartic acid (D), Glutamic acid (E), Asparagine (N), Glutamine (Q). *See also,* Creighton (1984) *Proteins, W.H.* Freeman and Company. In addition, individual substitutions, deletions or additions which alter, add or delete a single amino acid or a small percentage of amino acids in an encoded sequence are also "conservatively modified variations."

A "subsequence" refers to a sequence of nucleic acids or amino acids that comprise a part of a longer sequence of nucleic acids or amino acids (e.g., protein) respectively.

Nucleic acids are "elongated" when additional nucleotides (or other analogous molecules) are incorporated into the nucleic acid. Most commonly, this is performed with a polymerase (e.g., a DNA polymerase), e.g., a polymerase which adds sequences at the 3' terminus of the nucleic acid.

Two nucleic acids are "recombined" when sequences from each of the two nucleic acids are combined in a progeny nucleic acid. Two sequences are "directly" recombined when both of the nucleic acids are substrates for recombination. Two sequences are "indirectly recombined" when the sequences are recombined using an intermediate such as a cross-over oligonucleotide. For indirect recombination, no more than one of the sequences is an actual substrate for recombination, and in some cases, neither sequence is a substrate for recombination.

A "specific binding affinity" between two molecules, for example, a ligand and a receptor, means a preferential binding of one molecule for another in a mixture of molecules. The binding of the molecules can be considered specific if the binding affinity is about 1 x 10^4 M^{-1} to about 1 x 10^6 M^{-1} or greater.

Transformation: a process for introducing heterologous DNA into a host cell or organism.

"Transformed," "transgenic," and "recombinant" refer to a host organism such as a bacterium or a plant into which a heterologous nucleic acid molecule has been introduced. The nucleic acid molecule can be stably integrated into the genome of the host or the nucleic acid molecule can also be present as an extrachromosomal molecule. Such an extrachromosomal molecule can be auto-replicating. Transformed cells, tissues, or plants are understood to encompass not only the end product of a transformation process, but also transgenic progeny thereof. A "non-transformed," "non-transgenic," or "non-recombinant" host refers to a wild-type organism, e.g., a bacterium or plant, which does not contain the heterologous nucleic acid molecule.

The present invention addresses the aforementioned needs by providing several homologues of the *Arabidopsis NIM1* gene from additional species of plants. In particular, the present invention concerns the isolation of *Nicotiana tabacum* (tobacco), *Lycopersicon esculentum* (tomato), *Brassica napus* (oilseed rape), *Arabidopsis thaliana*, *Beta vulgaris* (sugarbeet), *Helianthus annuus* (sunflower), and *Solanum tuberosum* (potato) homologues of the *NIM1* gene, which encode proteins believed to be involved in the signal transduction cascade responsive to biological and chemical inducers that lead to systemic acquired resistance in plants.

Hence, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that encodes SEQ ID NO:2, 4, 6, 8, 16, 18, 20, 30, 32, 34, 36, 38, 40, 42, 44, 46, 48, 50, 52, 54, 56, 58, 62, 64, 66, 68, 70, 72, or 74.

In another embodiment, the present invention is directed to an isolated nucleic acid molecule comprising SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

In a further embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that comprises an at least 20, 25, 30, 35, 40, 45, or 50 (preferably 20) consecutive base pair portion identical in sequence to an at least 20, 25, 30, 35, 40, 45, or 50 (preferably 20) consecutive base pair portion of SEQ ID NO:1, 3, 5, 7, 15,

17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

In still another embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from a *Lycopersicon esculentum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, SEQ ID NO:26 and 28, or SEQ ID NO:59 and 60.

In yet another embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from a *Beta vulgaris* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:22 and 24 or SEQ ID NO:26 and 28.

In a further embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from a *Helianthus annuus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:26 and 28.

In another embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from a *Solanum tuberosum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:21 and 24, SEQ ID NO:21 and 23, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28.

In a further embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from a *Brassica napus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10 or SEQ ID NO:26 and 28.

In yet another embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from an *Arabidopsis thaliana* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:13 and 14, SEQ ID NO:21 and 24, or SEQ ID NO:22 and 24.

In a further embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from an *Nicotiana tabacum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:11 and 12, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28; or

In a further embodiment, the present invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that can be amplified from an plant DNA library using the polymerase chain reaction with a pair of primers comprising the first 20 nucleotides and the reverse complement of the last 20 nucleotides of the coding sequence (CDS) of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

The present invention also encompasses a chimeric gene comprising a promoter active in plants operatively linked to a *NIM1* homologue coding sequence of the present invention, a recombinant vector comprising such a chimeric gene, wherein the vector is capable of being stably transformed into a host, as well as a host stably transformed with such a vector. Preferably, the host is a plant such as one of the following agronomically important crops: rice, wheat, barley, rye, canola, sugarcane, corn, potato, carrot, sweet potato, sugar beet, bean, pea, chicory, lettuce, cabbage, cauliflower, broccoli, turnip, radish, spinach, asparagus, onion, garlic, eggplant, pepper, celery, squash, pumpkin, cucumber, apple, pear, quince, melon, plum, cherry, peach, nectarine, apricot, strawberry, grape, raspberry, blackberry, pineapple, avocado, papaya, mango, banana, soybean, tobacco, tomato, sorghum, and sugarcane. The present invention also encompasses seed from a plant of the invention.

Further, the present invention is directed to a method of increasing SAR gene expression in a plant by expressing in the plant a chimeric gene that itself comprises a promoter active in plants operatively linked to a *NIM1* homologue coding sequence of the present invention, wherein the encoded protein is expressed in the transformed plant at higher levels than in a wild type plant.

In addition, the present invention is directed to a method of enhancing disease resistance in a plant by expressing in the plant a chimeric gene that itself comprises a promoter active in plants operatively linked to a *NIM1* homologue coding sequence of the present invention, wherein the encoded protein is expressed in the transformed plant at higher levels than in a wild type plant.

Further, the present invention is directed to a PCR primer selected from the group consisting of SEQ ID NO:9-14, 21-28, 59, and 60.

The present invention also encompasses a method for isolating a *NIM1* homologue involved in the signal transduction cascade leading to systemic acquired resistance in plants comprising amplifying a DNA molecule from a plant DNA library using the polymerase chain reaction with a pair of primers corresponding to the first 20 nucleotides and the

reverse complement of the last 20 nucleotides of the coding sequence (CDS) of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73 or with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:11 and 12, SEQ ID NO:13 and 14, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:21 and 23, SEQ ID NO:25 and 28, SEQ ID NO:26 and 28, or SEQ ID NO:59 and 60. In a preferred embodiment, the plant DNA library is a *Nicotiana tabacum* (tobacco), *Lycopersicon esculentum* (tomato), *Brassica napus* (oilseed rape), *Arabidopsis thaliana*, *Beta vulgaris* (sugarbeet), *Helianthus annuus* (sunflower), or *Solanum tuberosum* (potato) DNA library.

Northern data on several of the *NIM1* homologues described herein indicates constitutive expression or BTH-inducibility. The homologues of the *NIM1* gene described herein are predicted to encode proteins involved in the signal transduction cascade responsive to biological and chemical inducers, which leads to systemic acquired resistance in plants. The present invention also concerns the transgenic expression of such *NIM1* homologues in plants to increase SAR gene expression and enhance disease resistance.

The DNA sequences of the invention can be isolated using the techniques described in the examples below, or by PCR using the sequences set forth in the sequence listing as the basis for constructing PCR primers. For example, oligonucleotides having the sequence of approximately the first and last 20-25 consecutive nucleotides of SEQ ID NO:7 (e.g., nucleotides 1-20 and 1742-1761 of SEQ ID NO:7) can be used as PCR primers to amplify the cDNA sequence (SEQ ID NO:7) directly from a cDNA library from the source plant (*Arabidopsis thaliana*). The other DNA sequences of the invention can likewise be amplified by PCR from cDNA or genomic DNA libraries of the respective plants using the ends of the DNA sequences set forth in the sequence listing as the basis for PCR primers.

The transgenic expression of the *NIM1* homologues of the invention in plants is predicted to result in immunity to a wide array of plant pathogens, which include, but are not limited to viruses or viroids, e.g. tobacco or cucumber mosaic virus, ringspot virus or necrosis virus, pelargonium leaf curl virus, red clover mottle virus, tomato bushy stunt virus, and like viruses; fungi, e.g. oomycetes such as *Phythophthora parasitica* and *Peronospora tabacina*; bacteria, e.g. *Pseudomonas syringae* and *Pseudomonas tabaci*; insects such as aphids, e.g. *Myzus persicae*; and lepidoptera, e.g., *Heliothus spp.*; and nematodes, e.g., *Meloidogyne incognita*. The vectors and methods of the invention are useful against a number of disease organisms of maize including but not limited to downy mildews such as *Sclerophthora macrospora*, *Sclerophthora rayissiae*, *Sclerospora graminicola*,

Peronosclerospora sorghi, *Peronosclerospora philippinensis*, *Peronosclerospora sacchari* and *Peronosclerospora maydis*; rusts such as *Puccinia sorphi*, *Puccinia polysora* and *Physopella zeae*; other fungi such as *Cercospora zeae-maydis*, *Colletotrichum graminicola*, *Fusarium monoliforme*, *Gibberella zeae*, *Exserohilum turcicum*, *Kabatiellu zeae*, *Erysiphe graminis*, *Septoria* and *Bipolaris maydis*; and bacteria such as *Erwinia stewartii*.

The methods of the present invention can be utilized to confer disease resistance to a wide variety of plants, including gymnosperms, monocots, and dicots. Although disease resistance can be conferred upon any plants falling within these broad classes, it is particularly useful in agronomically important crop plants, such as rice, wheat, barley, rye, rape, corn, potato, carrot, sweet potato, sugar beet, bean, pea, chicory, lettuce, cabbage, cauliflower, broccoli, turnip, radish, spinach, asparagus, onion, garlic, eggplant, pepper, celery, carrot, squash, pumpkin, zucchini, cucumber, apple, pear, quince, melon, plum, cherry, peach, nectarine, apricot, strawberry, grape, raspberry, blackberry, pineapple, avocado, papaya, mango, banana, soybean, tobacco, tomato, sorghum and sugarcane.

A *NIM1* homologue coding sequence of the present invention may be inserted into an expression cassette designed for plants to construct a chimeric gene according to the invention using standard genetic engineering techniques. The choice of specific regulatory sequences such as promoter, signal sequence, 5' and 3' untranslated sequences, and enhancer appropriate for the achieving the desired pattern and level of expression in the chosen plant host is within the level of skill of the routineer in the art. The resultant molecule, containing the individual elements linked in proper reading frame, may be inserted into a vector capable of being transformed into a host plant cell.

Examples of promoters capable of functioning in plants or plant cells (i.e., those capable of driving expression of associated coding sequences such as those coding for NIM1 homologues in plant cells) include the *Arabidopsis* and maize ubiquitin promoters; cauliflower mosaic virus (CaMV) 19S or 35S promoters and CaMV double promoters; rice actin promoters; PR-1 promoters from tobacco, *Arabidopsis*, or maize; nopaline synthase promoters; small subunit of ribulose bisphosphate carboxylase (ssuRUBISCO) promoters, and the like. Especially preferred is the *Arabidopsis* ubiquitin promoter. The promoters themselves may be modified to manipulate promoter strength to increase expression of the associated coding sequence in accordance with art-recognized procedures. Preferred promoters for use with the present invention are those that confer high level constitutive expression.

Signal or transit peptides may be fused to the *NIM1* homologue coding sequence in the chimeric DNA constructs of the invention to direct transport of the expressed protein to the desired site of action. Examples of signal peptides include those natively linked to the plant pathogenesis-related proteins, e.g. PR-1, PR-2, and the like. *See, e.g.,* Payne *et al.,* 1988. Examples of transit peptides include the chloroplast transit peptides such as those described in Von Heijne *et al.* (1991), *Mazur et al.* (1987), and Vorst *et al.* (1988); and mitochondrial transit peptides such as those described in Boutry *et al.* (1987). Also included are sequences that result in localization of the encoded protein to various cellular compartments such as the vacuole. See, for example, Neuhaus *et al.* (1991) and Chrispeels (1991).

The chimeric DNA construct(s) of the invention may contain multiple copies of a promoter or multiple copies of a *NIM1* homologue coding sequence of the present invention. In addition, the construct(s) may include coding sequences for markers and coding sequences for other peptides such as signal or transit peptides, each in proper reading frame with the other functional elements in the DNA molecule. The preparation of such constructs are within the ordinary level of skill in the art.

Useful markers include peptides providing herbicide, antibiotic or drug resistance, such as, for example, resistance to protoporphyrinogen oxidase inhibitors, hygromycin, kanamycin, G418, gentamycin, lincomycin, methotrexate, glyphosate, phosphinothricin, or the like. These markers can be used to select cells transformed with the chimeric DNA constructs of the invention from untransformed cells. Other useful markers are peptidic enzymes which can be easily detected by a visible reaction, for example a color reaction, for example luciferase, ß-glucuronidase, or ß-galactosidase.

Chimeric genes designed for plant expression such as those described herein can be introduced into the plant cell in a number of art-recognized ways. Those skilled in the art will appreciate that the choice of method might depend on the type of plant (i.e. monocot or dicot) and/or organelle (i.e. nucleus, chloroplast, mitochondria) targeted for transformation. Suitable methods of transforming plant cells include microinjection (Crossway *et al.,* 1986), electroporation (Riggs *et al.,* 1986), *Agrobacterium* mediated transformation (Hinchee *et al.,* 1988; Ishida *et al.,* 1996), direct gene transfer (Paszkowski *et al.,* 1984; Hayashimoto *et al.,* 1990), and ballistic particle acceleration using devices available from Agracetus, Inc., Madison, Wisconsin and Dupont, Inc., Wilmington, Delaware (*see, for example,* U.S. Patent 4,945,050; and McCabe *et al.,* 1988). *See also,* Weissinger *et al.* (1988); Sanford *et al.* (1987) (onion); Christou *et al.* (1988) (soybean); McCabe *et al.* (1988) (soybean); Datta *et*

al. (1990) (rice); Klein *et al.* (1988) (maize); Klein *et al.* (1988) (maize); Klein *et al.* (1988) (maize); Fromm *et al.* (1990); and Gordon-Kamm *et al.* (1990) (maize); Svab *et al.* (1990) (tobacco chloroplasts); Gordon-Kamm *et al.* (1993) (maize); Shimamoto *et al.* (1989) (rice); Christou *et al.* (1991) (rice); Datta *et al.* (1990) (rice); European Patent Application EP 0 332 581 (orchardgrass and other *Pooideae*); Vasil *et al.* (1993) (wheat); Weeks *et al.* (1993) (wheat); Wan *et al.* (1994) (barley); Jahne *et al.* (1994) (barley); Umbeck *et al.* (1987) (cotton); Casas *et al.* (1993) (sorghum); Somers *et al.* (1992) (oats); Torbert *et al.* (1995) (oats); Weeks *et al.*,(1993) (wheat); WO 94/13822 (wheat); and Nehra *et al.* (1994) (wheat). A particularly preferred set of embodiments for the introduction of recombinant DNA molecules into maize by microprojectile bombardment can be found in Koziel *et al.* (1993); Hill *et al.* (1995) and Koziel *et al.* (1996). An additional preferred embodiment is the protoplast transformation method for maize as disclosed in EP 0 292 435.

Once a chimeric gene comprising a *NIM1* homologue coding sequence has been transformed into a particular plant species, it may be propagated in that species or moved into other varieties of the same species, particularly including commercial varieties, using traditional breeding techniques. Particularly preferred plants of the invention include the agronomically important crops listed above. The genetic properties engineered into the transgenic seeds and plants described above are passed on by sexual reproduction and can thus be maintained and propagated in progeny plants.

EXAMPLES

The invention is illustrated in further detail by the following detailed procedures, preparations, and examples. The examples are for illustration only, and are not to be construed as limiting the scope of the present invention. Standard recombinant DNA and molecular cloning techniques used here are well known in the art and are described by Sambrook, *et al.*, 1989; by T.J. Silhavy, M.L. Berman, and L.W. Enquist, 1984; and by Ausubel, F.M. *et al.*, 1987.

I. Isolation of Homologues of the *Arabidopsis NIM1* Gene

Example 1: Isolation of a *NIM1* Homologue from *Nicotiana tabacum*

Plasmid DNA from a mass excision of phage from a tobacco cDNA library is used as a template for PCR using the following primer pairs: 5'-AGATTATTGTCAAGTCTAATG-3' (SEQ ID NO:9) + 5'-TTCCATGTACCTTTGCTTC-3' (SEQ ID NO:10), and 5'-GCGGATCCATGGATAATAGTAGG-3' (SEQ ID NO:11) + 5'-GCGGATCCTATTTCCTAAAAGGG-3' (SEQ ID NO:12). Cycling conditions are preferably 94 degrees for one minute, 40 degrees for one minute, and 72 degrees for 1.5 minutes, and the reaction is preferably carried out for 40 cycles. PCR products are run out on agarose gels, excised, and cloned into pCRII-TOPO (Invitrogen).

The full-length cDNA sequence of this tobacco *NIM1* homologue is shown in SEQ ID NO:1, and the protein encoded by this cDNA sequence is shown in SEQ ID NO:2. A tobacco *NIM1* homologue comprising SEQ ID NO:1 has been deposited as pNOV1206 with the NRRL (Agricultural Research Service, Patent Culture Collection, Northern Regional Research Center, 1815 North University Street, Peoria, Illinois 61604, U.S.A) on August 17, 1998, and assigned accession no. NRRL B-30051.

Example 2: Isolation of a *NIM1* Homologue from *Lycopersicon esculentum*

Phagemids are excised from λ ZAPII cDNA libraries of tomato using a protocol from Stratagene. Phagemids (plasmids) are mass-transformed into *E. coli* XL1-Blue in 10 pools of about 80,000 clones each and DNA is extracted from these pools. The pools are screened by PCR for the presence of *NIM1* homologues by PCR using the following

primers: 5'-AGATTATTGTCAAGTCTAATG-3' (SEQ ID NO:9) and 5'-TTCCATGTACCTTTGCTTC-3' (SEQ ID NO:10).

Sequences amplified from the pools are confirmed to contain *NIM1* homologues by cloning the PCR-amplified DNA fragment and sequencing. Pools are made successively smaller and screened by PCR using the same primers mentioned above for the presence of the *NIM1* homologues until a single clone containing the homologue is obtained. In the event that the cDNA clone contains a partial gene missing the 5' end, 5' RACE (Rapid Amplification of cDNA Ends) is used to obtain the full-length sequence of the gene.

The full-length cDNA sequence of this tomato *NIM1* homologue is shown in SEQ ID NO:3, and the protein encoded by this cDNA sequence is shown in SEQ ID NO:4. A tomato *NIM1* homologue comprising SEQ ID NO:3 has been deposited as pNOV1204 with the NRRL (Agricultural Research Service, Patent Culture Collection, Northern Regional Research Center, 1815 North University Street, Peoria, Illinois 61604, U.S.A) on August 17, 1998, and assigned accession no. NRRL B-30050.

Example 3: Isolation of a *NIM1* Homologue from *Brassica napus*

Phagemids are excised from λ ZAPII cDNA libraries of *Brassica napus* using a protocol from Stratagene. Phagemids (plasmids) are mass-transformed into *E. coli* XL1-Blue in 10 pools of about 80,000 clones each and DNA is extracted from these pools. The pools are screened by PCR for the presence of *NIM1* homologues by PCR using the following primers: 5'-AGATTATTGTCAAGTCTAATG-3' (SEQ ID NO:9) and 5'-TTCCATGTACCTTTGCTTC-3' (SEQ ID NO:10).

Sequences amplified from the pools are confirmed to contain *NIM1* homologues by cloning the PCR-amplified DNA fragment and sequencing. Pools are made successively smaller and screened by PCR using the same primers mentioned above for the presence of the *NIM1* homologues until a single clone containing the homologue is obtained. In the event that the cDNA clone contains a partial gene, missing the 5' end, 5' RACE (Rapid Amplification of cDNA Ends) is used to obtain the full-length sequence of the gene.

A partial cDNA sequence of this *Brassica napus NIM1* homologue is shown in SEQ ID NO:5, and the protein encoded by this cDNA sequence is shown in SEQ ID NO:6. A *Brassica napus NIM1* homologue comprising SEQ ID NO:5 has been deposited as pNOV1203 with the NRRL (Agricultural Research Service, Patent Culture Collection,

Northern Regional Research Center, 1815 North University Street, Peoria, Illinois 61604, U.S.A) on August 17, 1998, and assigned accession no. NRRL B-30049.

Example 4: Isolation of a *NIM1* Homologue from *Arabidopsis thaliana*

BLAST searches using the *Arabidopsis* or tomato NIM1 amino acid sequences as queries detect GenBank entry B26306, which contains *Arabidopsis* genomic sequence from the Bacterial Artificial Chromosome (BAC) F18D8. Part of the BAC sequence is predicted to encode a protein with significant similarity (47% amino acid identity) to NIM1. The following primers are designed to regions of the F18D8 sequence: 5'-TCAAGGCCTTGGATTCAGATG-3' (SEQ ID NO:13) and 5'-ATTAACTGCGCTACGTCCGTC-3' (SEQ ID NO:14).

The primers are used in a PCR reaction with DNA from a pFL61-based *Arabidopsis* cDNA library as a template. Preferable cycling conditions are 94 degrees for 30 seconds, 53 degrees for 30 seconds, 72 degrees for 30 seconds. The reaction is preferably run for 40 cycles. A PCR product of the predicted size (290 base pairs) is detected, and the cDNA clone corresponding to the F18D8 primers is purified from the cDNA library by sequential purification by passage of increasingly smaller amounts of the library through *E. coli* and re-diagnosis of the presence of the clone by PCR. Ultimately, a single positive clone is obtained and sequenced. The sequence of the clone confirms the presence of an open reading frame with significant homology to NIM1.

A full-length cDNA sequence of this *Arabidopsis thaliana NIM1* homologue is shown in SEQ ID NO:7, and the protein encoded by this cDNA sequence is shown in SEQ ID NO:8. An *Arabidopsis thaliana NIM1* homologue comprising SEQ ID NO:7 has been deposited as *AtNMLc5* in *E. coli* with the NRRL (Agricultural Research Service, Patent Culture Collection, Northern Regional Research Center, 1815 North University Street, Peoria, Illinois 61604, U.S.A) on May 25, 1999, and assigned accession no. NRRL B-30139.

Example 5: Design of Degenerate Primers

In addition to the *NIM1* gene (Ryals *et al.*, 1997) and the *NIM*-like gene described above in Example 4 (*AtNMLc5* - SEQ ID NO:7), *Arabidopsis thaliana* contains three other *NIM*-like (*NML*) genomic sequences: *AtNMLc2* (SEQ ID NO:15), *AtNMLc4-1* (SEQ ID

NO:17), and *AtNMLc4-2* (SEQ ID NO:19), where *c[#]* stands for the chromosome number on which the particular *NML* gene is located. Using the GCG Seqweb multiple sequence alignment program (Pretty, Wisconsin Genetics Computer Group), the *NIM1* sequences from *Arabidopsis thaliana* (Ryals *et al.*, 1997), *Nicotiana tabacum* (Example 1 - SEQ ID NO:1), and *Lycopersicon esculentum* (Example 2 - SEQ ID NO:3), as well as the *NML* sequences from *Arabidopsis thaliana* (SEQ ID NO:7, 15, 17, and 19) are aligned. Based on this alignment, three regions emerge with sufficient conservation to design degenerate PCR primers for PCR amplification of *NIM1* homologues from other crop species, including sugarbeet, sunflower, potato, and canola. The primers designed from these conserved regions are listed below in Table 1. The NIM 1(A-D) primers are designed using a lineup with only the *NIM1* genes from *Arabidopsis thaliana* (Ryals *et al.*, 1997), *Nicotiana tabacum* (Example 1 - SEQ ID NO:1), and *Lycopersicon esculentum* (Example 2 - SEQ ID NO:3). The NIM 2(A-D) primers are designed using a lineup with these three sequences in addition to the four *NML* sequences from *Arabidopsis thaliana* (SEQ ID NO:7, 15, 17, and 19). Primers are preferably synthesized by Genosys Biotechnologies, Inc. (The Woodlands, Texas). Positions of degeneracy are indicated in Table 1 by the notation of more than one base at a single site in the oligonucleotide. "Orientation" designates whether the primer is directed towards the 3' end (Downstream) or the 5' end (Upstream) of the cDNA.

Table 1: Degenerate Primers

Primer	Sequence (5' to 3')	SEQ ID NO:	Orientation
NIM 1A	GAGATTATTGTCAAGTCTAATGTAGATA T T	SEQ ID NO:21	Downstream
NIM 1B	ACTGGACTCGGATGATATTGAATTA T T T T G G	SEQ ID NO:22	Downstream
NIM 1C	TAACTCAACATCATCAGAATCAAATGC T T C G C G	SEQ ID NO:23	Upstream
NIM 1D	GTTGAGCAAGAGCAACTCTATTTTCAAG T C CC G T	SEQ ID NO:24	Upstream
NIM 2A	TGCATAGAAATAATTGTGAAGTCTAATGTAGA T G TG C G T	SEQ ID NO:25	Downstream
NIM 2B	GGCACTGGACTCAGATGATGTTGAACT T T T GT	SEQ ID NO:26	Downstream
NIM 2C	AACTCAACATCATCAGAATCCAATGCC GT T G G	SEQ ID NO:27	Upstream
NIM 2D	AGTTGAGCAAGGCCAACTCGATTTTCAAAAT T C A T GG T	SEQ ID NO:28	Upstream

Example 6: PCR Amplification of *NIM*-like DNA Fragments From Crop Species

NIM-like DNA fragments are amplified from *Arabidopsis*, tomato, tobacco, sugarbeet, sunflower, potato, and canola, using either genomic DNA or cDNA as templates. The primer combinations used, along with the expected fragment sizes, are listed below in Table 2.

Table 2: Primer combinations and DNA fragment sizes

Left Primer	Right Primer	Fragment Size (bp)
NIM 1A	NIM 1D	669
NIM 1A	NIM 1C	195
NIM 1B	NIM 1D	499
NIM 2A	NIM 2D	676
NIM 2A	NIM 2C	200
NIM 2B	NIM 2D	503

Degenerate primer PCR is preferably performed with Ready-To-Go PCR Beads (Amersham, Piscataway, NJ) in a GeneAmp PCR System 9700 (PE Applied Biosystems, Foster City, CA). 20 to 40 ng of genomic DNA or 5 to 10 ng of cDNA is used in each reaction, with each primer at a final concentration of 0.8 µM. Preferable cycling parameters are as follows: 94°C for 1 minute; 3 cycles of [94°C for 30 seconds; 37°C for 30 seconds; 72°C for 2 minutes]; 35 cycles of [94°C for 30 seconds; 60°C for 30 seconds; 72°C for 2 minutes]; 72°C for 7 minutes; 4°C hold. Reaction products are analyzed on 2% agarose gels and DNA fragments of the appropriate size are excised. DNA fragments are isolated from agarose bands using, for example, the Geneclean III Kit (BIO 101, Inc., Carlsbad, CA) and cloned using, for example, the TOPO TA Cloning Kit (Invitrogen Corporation, Carlsbad, CA). Plasmids are isolated using, for example, the CONCERT Rapid Plasmid Miniprep System (Life Technologies, Inc., Rockville, MD) and sequenced by standard protocols.

NIM-like DNA fragments are obtained from all plant species attempted, and in many cases multiple, unique *NIM*-like sequences are isolated. Table 3 and Figure 2 detail the *NIM*-like fragments that are isolated.

Table 3: *NIM*-like PCR fragments

Species	Successful Primer Pairs	PCR Template	Unique Clones	SEQ ID NO:
Arabidopsis	1A/1D; 1B/1D	Genomic DNA	One	
Tobacco	1A/1D; 1B/1D; 2A/2D; 2B/2D	cDNA	Four	SEQ ID NO: 29, 31, 33, and 35
Tomato	1A/1D; 1B/1D; 2A/2D; 2B/2D	Genomic DNA, cDNA	One	SEQ ID NO: 37
Sugarbeet	1B/1D; 2B/2D	Genomic DNA, cDNA	One	SEQ ID NO: 39
Sunflower	2B/2D	cDNA	Two	SEQ ID NO: 41 and 43
Potato	1A/1D; 1A/1C; 1B/1D; 2A/2D; 2B/2D	cDNA	Three	SEQ ID NO: 45, 47, and 49
Canola	2B/2D	cDNA	Four	SEQ ID NO: 51, 53, 55, and 57

Based on these results, the degenerate primer PCR described above can amplify *NIM*-like fragments from a wide variety of plant species. In particular, the primer combination of NIM 2B/NIM 2D is successful with cDNA as a template from all species attempted. The use of Ready-To-Go PCR Beads is especially preferably for obtaining products. In addition, using cDNA as a template is preferable for all samples except *Arabidopsis*, tomato and sugarbeet, where genomic DNA is sufficient.

Example 7: Additional Degenerate Primers

A new pair of degenerate primers is designed based on a sequence alignment of the four tobacco fragments (SEQ ID NO: 29, 31, 33, and 35) and the tomato sequence (SEQ ID NO: 37) for use in determining whether tomato also contains similar *NIM*-like sequences that are not amplified with the degenerate primers listed in Table 1. The primers designed from these fragments are listed below in Table 3 and are preferably synthesized by Genosys Biotechnologies, Inc. (The Woodlands, Texas). Positions of degeneracy are indicated in Table 3 by the notation of more than one base at a single site in the

oligonucleotide. "Orientation" designates whether the primer is directed towards the 3' end (Downstream) or the 5' end (Upstream) of the cDNA.

Table 4: Additional degenerate primers

Primer	Sequence (5' TO 3')	SEQ ID NO:	Orientation
NIM 3A	TAGATGAAGCATACGCTCTCCACTATGCTGT T C T T T	SEQ ID NO:59	Downstream
NIM 3B	GGCTCCTTACGCATGGCAGCAACATGAAGGAC T C T TG C	SEQ ID NO:60	Upstream

Degenerate primer PCR is performed as described above using tomato cDNA, and potential products are cloned and sequenced. The sequence analysis reveals two classes of *NIM*-like fragments: the first is identical to the tomato sequence shown in SEQ ID NO: 37, and the second is unique in tomato and 88% identical to the tobacco sequences shown in SEQ ID NO:31 and 33. The sequence of this new tomato sequence is presented in SEQ ID NO:61.

Example 8: Full-length *NIM*-like cDNA's

Corresponding cDNA sequences upstream and downstream from *NIM*-like PCR fragments are preferably obtained by RACE PCR using the SMART RACE cDNA Amplification Kit (Clontech, Palo Alto, CA). Preferably, at least three independent RACE products are sequenced for each 5'- or 3'-end in order to eliminate PCR errors. Resulting full-length cDNA sequences for Sugarbeet, Sunflower B, and Tobacco B *NIM1* homologues, which correspond to the *NIM*-like PCR fragments shown in SEQ ID NO:39, 43, and 31 are presented as SEQ ID NO:63, 65, and 73 respectively.

NIM-like *Arabidopsis thaliana* cDNA's corresponding to the *NIM*-like genomic sequences *AtNMLc2* (SEQ ID NO:15), *AtNMLc4-1* (SEQ ID NO:17), and *AtNMLc4-2* (SEQ ID NO:19), are preferably cloned by RT-PCR. Total RNA from *Arabidopsis thaliana* is reverse transcribed using oligo dT primer. The resulting first strand cDNA is amplified by PCR using specific sense and antisense oligonucleotide primers designed based on the 5' and 3' ends of the coding region of each genomic sequence (SEQ ID NO:15, 17, and 19). PCR fragments of the predicted sizes are cloned into a vector and sequenced to confirm that these cDNA clones correspond to the *NIM*-like genomic sequences. A cDNA sequence corresponding to the *NIM*-like genomic sequence *AtNMLc2* (SEQ ID NO:15) is presented as

SEQ ID NO:67; a full-length cDNA sequence corresponding to the *NIM*-like genomic sequence *AtNMLc4-1* (SEQ ID NO:17) is presented as SEQ ID NO:69; and a full-length cDNA sequence corresponding to the *NIM*-like genomic sequence *AtNMLc4-2* (SEQ ID NO:19) is presented as SEQ ID NO:71.

Example 9: Northern Analysis

Northern data shows that expression of the sugarbeet *NIM*-like clone (SEQ ID NO:39 and 63) increases three to seven fold after 100µM or 300 µM BTH (benzo(1,2,3)thiadiazole-7-carbothioic acid *S*-methyl ester) treatment. Also, Northern data shows that expression of the Sunflower A *NIM*-like clone (SEQ ID NO:41) is constitutive. Furthermore, Northern data shows that expression of the Sunflower B *NIM*-like clone (SEQ ID NO:43 and 65) increases two fold after 100µM or 300 µM BTH treatment.

II. Expression of the Gene Sequences of the Invention In Plants

A *NIM1* homologue of the present invention can be incorporated into plant cells using conventional recombinant DNA technology. Generally, this involves inserting a coding sequence of the invention into an expression system to which the coding sequence is heterologous (i.e., not normally present) using standard cloning procedures known in the art. The vector contains the necessary elements for the transcription and translation of the inserted protein-coding sequences. A large number of vector systems known in the art can be used, such as plasmids, bacteriophage viruses and other modified viruses. Suitable vectors include, but are not limited to, viral vectors such as lambda vector systems λgtl1, λgtl0 and Charon 4; plasmid vectors such as pBI121, pBR322, pACYC177, pACYC184, pAR series, pKK223-3, pUC8, pUC9, pUC18, pUC19, pLG339, pRK290, pKC37, pKC101, pCDNAII; and other similar systems. The components of the expression system may also be modified to increase expression. For example, truncated sequences, nucleotide substitutions or other modifications may be employed. The expression systems described herein can be used to transform virtually any crop plant cell under suitable conditions. Transformed cells can be regenerated into whole plants such that the *NIM1* homologue increases SAR gene expression and enhances disease resistance in the transgenic plants.

Example 10: Construction of Plant Expression Cassettes

Coding sequences intended for expression in transgenic plants are first assembled in expression cassettes behind a suitable promoter expressible in plants. The expression cassettes may also comprise any further sequences required or selected for the expression of the transgene. Such sequences include, but are not restricted to, transcription terminators, extraneous sequences to enhance expression such as introns, vital sequences, and sequences intended for the targeting of the gene product to specific organelles and cell compartments. These expression cassettes can then be easily transferred to the plant transformation vectors described below. The following is a description of various components of typical expression cassettes.

1. Promoters

The selection of the promoter used in expression cassettes will determine the spatial and temporal expression pattern of the transgene in the transgenic plant. Selected promoters will express transgenes in specific cell types (such as leaf epidermal cells, mesophyll cells, root cortex cells) or in specific tissues or organs (roots, leaves or flowers, for example) and the selection will reflect the desired location of accumulation of the gene product. Alternatively, the selected promoter may drive expression of the gene under various inducing conditions. Promoters vary in their strength, i.e., ability to promote transcription. Depending upon the host cell system utilized, any one of a number of suitable promoters can be used, including the gene's native promoter. The following are non-limiting examples of promoters that may be used in expression cassettes.

a. Constitutive Expression, the Ubiquitin Promoter:

Ubiquitin is a gene product known to accumulate in many cell types and its promoter has been cloned from several species for use in transgenic plants (*e.g.* sunflower - Binet *et al.*,1991; maize - Christensen *et al.*, 1989; and *Arabidopsis* - Norris *et al.*, 1993). The maize ubiquitin promoter has been developed in transgenic monocot systems and its sequence and vectors constructed for monocot transformation are disclosed in the patent publication EP 0 342 926 (to Lubrizol). Taylor *et al.* (1993) describe a vector (pAHC25) that comprises the maize ubiquitin promoter and first intron and its high activity in cell suspensions of numerous monocotyledons when introduced via microprojectile

bombardment. The *Arabidopsis* ubiquitin promoter is especially preferred for use with the *NIM1* homologues of the present invention. The ubiquitin promoter is suitable for gene expression in transgenic plants, both monocotyledons and dicotyledons. Suitable vectors are derivatives of pAHC25 or any of the transformation vectors described in this application, modified by the introduction of the appropriate ubiquitin promoter and/or intron sequences.

b. Constitutive Expression, the CaMV 35S Promoter:

Construction of the plasmid pCGN1761 is described in the published patent application EP 0 392 225 (Example 23). pCGN1761 contains the "double" CaMV 35S promoter and the *tml* transcriptional terminator with a unique *EcoRI* site between the promoter and the terminator and has a pUC-type backbone. A derivative of pCGN1761 is constructed which has a modified polylinker which includes *NotI* and *XhoI* sites in addition to the existing *EcoRI* site. This derivative is designated pCGN1761ENX. pCGN1761ENX is useful for the cloning of cDNA sequences or coding sequences (including microbial ORF sequences) within its polylinker for the purpose of their expression under the control of the 35S promoter in transgenic plants. The entire 35S promoter-coding sequence-*tml* terminator cassette of such a construction can be excised by *HindIII, SphI, SalI,* and *XbaI* sites 5' to the promoter and *XbaI, BamHI* and *BglI* sites 3' to the terminator for transfer to transformation vectors such as those described below. Furthermore, the double 35S promoter fragment can be removed by 5' excision with *HindIII, SphI, SalI, XbaI,* or *PstI,* and 3' excision with any of the polylinker restriction sites (*EcoRI, NotI* or *XhoI*) for replacement with another promoter. If desired, modifications around the cloning sites can be made by the introduction of sequences that may enhance translation. This is particularly useful when overexpression is desired. For example, pCGN1761ENX may be modified by optimization of the translational initiation site as described in Example 37 of U.S. Patent No. 5,639,949.

c. Constitutive Expression, the Actin Promoter:

Several isoforms of actin are known to be expressed in most cell types and consequently the actin promoter is a good choice for a constitutive promoter. In particular, the promoter from the rice *Actl* gene has been cloned and characterized (McElroy *et al.*, 1990). A 1.3kb fragment of the promoter was found to contain all the regulatory elements required for expression in rice protoplasts. Furthermore, numerous expression vectors based on the *Actl* promoter have been constructed specifically for use in monocotyledons (McElroy *et al.*, 1991). These incorporate the *Actl*-intron 1, *Adhl* 5' flanking sequence and

Adh1-intron 1 (from the maize alcohol dehydrogenase gene) and sequence from the CaMV 35S promoter. Vectors showing highest expression were fusions of 35S and *Act1* intron or the *Act1* 5' flanking sequence and the *Act1* intron. Optimization of sequences around the initiating ATG (of the GUS reporter gene) also enhanced expression. The promoter expression cassettes described by McElroy *et al.* (1991) can be easily modified for gene expression and are particularly suitable for use in monocotyledonous hosts. For example, promoter-containing fragments is removed from the McElroy constructions and used to replace the double 35S promoter in pCGN1761ENX, which is then available for the insertion of specific gene sequences. The fusion genes thus constructed can then be transferred to appropriate transformation vectors. In a separate report, the rice *Act1* promoter with its first intron has also been found to direct high expression in cultured barley cells (Chibbar *et al.*, 1993).

d. Inducible Expression, the PR-1 Promoter:

The double 35S promoter in pCGN1761ENX may be replaced with any other promoter of choice that will result in suitably high expression levels. By way of example, one of the chemically regulatable promoters described in U.S. Patent No. 5,614,395 may replace the double 35S promoter. The promoter of choice is preferably excised from its source by restriction enzymes, but can alternatively be PCR-amplified using primers that carry appropriate terminal restriction sites. Should PCR-amplification be undertaken, then the promoter should be re-sequenced to check for amplification errors after the cloning of the amplified promoter in the target vector. The chemically/pathogen regulatable tobacco PR-1a promoter is cleaved from plasmid pCIB1004 (for construction, see example 21 of EP 0 332 104) and transferred to plasmid pCGN1761ENX (Uknes *et al.*, 1992). pCIB1004 is cleaved with *NcoI* and the resultant 3' overhang of the linearized fragment is rendered blunt by treatment with T4 DNA polymerase. The fragment is then cleaved with *HindIII* and the resultant PR-1a promoter-containing fragment is gel purified and cloned into pCGN1761ENX from which the double 35S promoter has been removed. This is done by cleavage with *XhoI* and blunting with T4 polymerase, followed by cleavage with *HindIII* and isolation of the larger vector-terminator containing fragment into which the pCIB1004 promoter fragment is cloned. This generates a pCGN1761ENX derivative with the PR-1a promoter and the *tml* terminator and an intervening polylinker with unique *EcoRI* and *NotI* sites. The selected coding sequence can be inserted into this vector, and the fusion products (*i.e.* promoter-gene-terminator) can subsequently be transferred to any selected

transformation vector, including those described *infra*. Various chemical regulators may be employed to induce expression of the selected coding sequence in the plants transformed according to the present invention, including the benzothiadiazole, isonicotinic acid, and salicylic acid compounds disclosed in U.S. Patent Nos. 5,523,311 and 5,614,395.

e. Inducible Expression, an Ethanol-Inducible Promoter:

A promoter inducible by certain alcohols or ketones, such as ethanol, may also be used to confer inducible expression of a coding sequence of the present invention. Such a promoter is for example the *alcA* gene promoter from *Aspergillus nidulans* (Caddick *et al.*, 1998). In *A. nidulans*, the *alcA* gene encodes alcohol dehydrogenase I, the expression of which is regulated by the AlcR transcription factors in presence of the chemical inducer. For the purposes of the present invention, the CAT coding sequences in plasmid palcA:CAT comprising a *alcA* gene promoter sequence fused to a minimal 35S promoter (Caddick *et al.*, 1998) are replaced by a coding sequence of the present invention to form an expression cassette having the coding sequence under the control of the *alcA* gene promoter. This is carried out using methods well known in the art.

f. Inducible Expression, a Glucocorticoid-Inducible Promoter:

Induction of expression of a NIM1 homologue of the present invention using systems based on steroid hormones is also contemplated. For example, a glucocorticoid-mediated induction system is used (Aoyama and Chua, 1997) and gene expression is induced by application of a glucocorticoid, for example a synthetic glucocorticoid, preferably dexamethasone, preferably at a concentration ranging from 0.1mM to 1mM, more preferably from 10mM to 100mM. For the purposes of the present invention, the luciferase gene sequences are replaced by a gene sequence encoding a NIM1 homologue to form an expression cassette having the gene sequence encoding a NIM1 homologue under the control of six copies of the GAL4 upstream activating sequences fused to the 35S minimal promoter. This is carried out using methods well known in the art. The trans-acting factor comprises the GAL4 DNA-binding domain (Keegan *et al.*, 1986) fused to the transactivating domain of the herpes viral protein VP16 (Triezenberg *et al.*, 1988) fused to the hormone-binding domain of the rat glucocorticoid receptor (Picard *et al.*, 1988). The expression of the fusion protein is controlled by any promoter suitable for expression in plants known in the art or described here. This expression cassette is also comprised in the plant comprising the gene sequence encoding a NIM1 homologue fused to the 6xGAL4/minimal promoter.

Thus, tissue- or organ-specificity of the fusion protein is achieved leading to inducible tissue- or organ-specificity of the NIM1 homologue.

g. Root Specific Expression:

Another pattern of gene expression is root expression. A suitable root promoter is described by de Framond (1991) and also in the published patent application EP 0 452 269. This promoter is transferred to a suitable vector such as pCGN1761ENX for the insertion of a selected gene and subsequent transfer of the entire promoter-gene-terminator cassette to a transformation vector of interest.

h. Wound-Inducible Promoters:

Wound-inducible promoters may also be suitable for gene expression. Numerous such promoters have been described (*e.g.* Xu *et al.*, 1993); Logemann *et al.*, 1989; Rohrmeier & Lehle, 1993; Firek *et al.*, 1993; Warner *et al.*, 1993) and all are suitable for use with the instant invention. Logemann *et al.* describe the 5' upstream sequences of the dicotyledonous potato *wunI* gene. Xu *et al.* show that a wound-inducible promoter from the dicotyledon potato (*pin2*) is active in the monocotyledon rice. Further, Rohrmeier & Lehle describe the cloning of the maize *WipI* cDNA which is wound induced and which can be used to isolate the cognate promoter using standard techniques. Similar, Firek *et al.* and Warner *et al.* have described a wound-induced gene from the monocotyledon *Asparagus officinalis,* which is expressed at local wound and pathogen invasion sites. Using cloning techniques well known in the art, these promoters can be transferred to suitable vectors, fused to the genes pertaining to this invention, and used to express these genes at the sites of plant wounding.

i. Pith-Preferred Expression:

Patent Application WO 93/07278 describes the isolation of the maize *trpA* gene, which is preferentially expressed in pith cells. The gene sequence and promoter extending up to -1726 bp from the start of transcription are presented. Using standard molecular biological techniques, this promoter, or parts thereof, can be transferred to a vector such as pCGN1761 where it can replace the 35S promoter and be used to drive the expression of a foreign gene in a pith-preferred manner. In fact, fragments containing the pith-preferred promoter or parts thereof can be transferred to any vector and modified for utility in transgenic plants.

j. Leaf-Specific Expression:

A maize gene encoding phosphoenol carboxylase (PEPC) has been described by Hudspeth & Grula (1989). Using standard molecular biological techniques the promoter for this gene can be used to drive the expression of any gene in a leaf-specific manner in transgenic plants.

k. Pollen-Specific Expression:

WO 93/07278 describes the isolation of the maize calcium-dependent protein kinase (CDPK) gene which is expressed in pollen cells. The gene sequence and promoter extend up to 1400 bp from the start of transcription. Using standard molecular biological techniques, this promoter or parts thereof, can be transferred to a vector such as pCGN1761 where it can replace the 35S promoter and be used to drive the expression of a NIM1 homologue of the present invention in a pollen-specific manner.

2. Transcriptional Terminators

A variety of transcriptional terminators are available for use in expression cassettes. These are responsible for the termination of transcription beyond the transgene and its correct polyadenylation. Appropriate transcriptional terminators are those that are known to function in plants and include the CaMV 35S terminator, the *tml* terminator, the nopaline synthase terminator and the pea *rbcS* E9 terminator. These can be used in both monocotyledons and dicotyledons. In addition, a gene's native transcription terminator may be used.

3. Sequences for the Enhancement or Regulation of Expression

Numerous sequences have been found to enhance gene expression from within the transcriptional unit and these sequences can be used in conjunction with the genes of this invention to increase their expression in transgenic plants.

Various intron sequences have been shown to enhance expression, particularly in monocotyledonous cells. For example, the introns of the maize *Adh1* gene have been found to significantly enhance the expression of the wild-type gene under its cognate promoter when introduced into maize cells. Intron 1 was found to be particularly effective and enhanced expression in fusion constructs with the chloramphenicol acetyltransferase gene (Callis *et al.*, 1987). In the same experimental system, the intron from the maize *bronze1*

gene had a similar effect in enhancing expression. Intron sequences have been routinely incorporated into plant transformation vectors, typically within the non-translated leader.

A number of non-translated leader sequences derived from viruses are also known to enhance expression, and these are particularly effective in dicotyledonous cells. Specifically, leader sequences from Tobacco Mosaic Virus (TMV, the "W-sequence"), Maize Chlorotic Mottle Virus (MCMV), and Alfalfa Mosaic Virus (AMV) have been shown to be effective in enhancing expression (*e.g.* Gallie *et al.*, 1987; Skuzeski *et al.*, 1990).

4. Targeting of the Gene Product Within the Cell

Various mechanisms for targeting gene products are known to exist in plants and the sequences controlling the functioning of these mechanisms have been characterized in some detail. For example, the targeting of gene products to the chloroplast is controlled by a signal sequence found at the amino terminal end of various proteins which is cleaved during chloroplast import to yield the mature protein (*e.g.* Comai *et al.*, 1988). These signal sequences can be fused to heterologous gene products to effect the import of heterologous products into the chloroplast (van den Broeck, *et al.*, 1985). DNA encoding for appropriate signal sequences can be isolated from the 5' end of the cDNAs encoding the RUBISCO protein, the CAB protein, the EPSP synthase enzyme, the GS2 protein and many other proteins which are known to be chloroplast localized. *See also*, the section entitled "Expression With Chloroplast Targeting" in Example 37 of U.S. Patent No. 5,639,949.

Other gene products are localized to other organelles such as the mitochondrion and the peroxisome (*e.g.* Unger *et al.*, 1989). The cDNAs encoding these products can also be manipulated to effect the targeting of heterologous gene products to these organelles. Examples of such sequences are the nuclear-encoded ATPases and specific aspartate amino transferase isoforms for mitochondria. Targeting cellular protein bodies has been described by Rogers *et al.* (1985).

In addition, sequences have been characterized which cause the targeting of gene products to other cell compartments. Amino terminal sequences are responsible for targeting to the ER, the apoplast, and extracellular secretion from aleurone cells (Koehler & Ho, 1990). Additionally, amino terminal sequences in conjunction with carboxy terminal sequences are responsible for vacuolar targeting of gene products (Shinshi *et al.*, 1990).

By the fusion of the appropriate targeting sequences described above to transgene sequences of interest it is possible to direct the transgene product to any organelle or cell compartment. For chloroplast targeting, for example, the chloroplast signal sequence from

the RUBISCO gene, the CAB gene, the EPSP synthase gene, or the GS2 gene is fused in frame to the amino terminal ATG of the transgene. The signal sequence selected should include the known cleavage site, and the fusion constructed should take into account any amino acids after the cleavage site which are required for cleavage. In some cases this requirement may be fulfilled by the addition of a small number of amino acids between the cleavage site and the transgene ATG or, alternatively, replacement of some amino acids within the transgene sequence. Fusions constructed for chloroplast import can be tested for efficacy of chloroplast uptake by *in vitro* translation of *in vitro* transcribed constructions followed by *in vitro* chloroplast uptake using techniques described by Bartlett *et al.* (1982) and Wasmann *et al.* (1986). These construction techniques are well known in the art and are equally applicable to mitochondria and peroxisomes.

The above-described mechanisms for cellular targeting can be utilized not only in conjunction with their cognate promoters, but also in conjunction with heterologous promoters so as to effect a specific cell-targeting goal under the transcriptional regulation of a promoter that has an expression pattern different to that of the promoter from which the targeting signal derives.

Example 11: Construction of Plant Transformation Vectors

Numerous transformation vectors available for plant transformation are known to those of ordinary skill in the plant transformation arts, and the genes pertinent to this invention can be used in conjunction with any such vectors. The selection of vector will depend upon the preferred transformation technique and the target species for transformation. For certain target species, different antibiotic or herbicide selection markers may be preferred. Selection markers used routinely in transformation include the *nptII* gene, which confers resistance to kanamycin and related antibiotics (Messing & Vierra, 1982; Bevan *et al.*, 1983), the *bar* gene, which confers resistance to the herbicide phosphinothricin (White *et al.*, 1990; Spencer *et al.*, 1990), the *hph* gene, which confers resistance to the antibiotic hygromycin (Blochinger & Diggelmann), and the *dhfr* gene, which confers resistance to methatrexate (Bourouis *et al.*, 1983), and the EPSPS gene, which confers resistance to glyphosate (U.S. Patent Nos. 4,940,935 and 5,188,642).

1. Vectors Suitable for *Agrobacterium* Transformation

Many vectors are available for transformation using *Agrobacterium tumefaciens*. These typically carry at least one T-DNA border sequence and include vectors such as pBIN19 (Bevan, Nucl. Acids Res. (1984)) and pXYZ. Below, the construction of two typical vectors suitable for *Agrobacterium* transformation is described.

a. pCIB200 and pCIB2001:

The binary vectors pcIB200 and pCIB2001 are used for the construction of recombinant vectors for use with *Agrobacterium* and are constructed in the following manner. pTJS75kan is created by *NarI* digestion of pTJS75 (Schmidhauser & Helinski, 1985) allowing excision of the tetracycline-resistance gene, followed by insertion of an *AccI* fragment from pUC4K carrying an NPTII (Messing & Vierra, 1982; Bevan *et al.*, 1983; McBride *et al.*, 1990). *XhoI* linkers are ligated to the *EcoRV* fragment of PCIB7 which contains the left and right T-DNA borders, a plant selectable *nos/nptII* chimeric gene and the pUC polylinker (Rothstein *et al.*, 1987), and the *XhoI*-digested fragment are cloned into *SalI*-digested pTJS75kan to create pCIB200 (see also EP 0 332 104, example 19). pCIB200 contains the following unique polylinker restriction sites: *EcoRI, SstI, KpnI, BglII, XbaI*, and *SalI*. pCIB2001 is a derivative of pCIB200 created by the insertion into the polylinker of additional restriction sites. Unique restriction sites in the polylinker of pCIB2001 are *EcoRI, SstI, KpnI, BglII, XbaI, SalI, MluI, BclI, AvrII, ApaI, HpaI*, and *StuI*. pCIB2001, in addition to containing these unique restriction sites also has plant and bacterial kanamycin selection, left and right T-DNA borders for *Agrobacterium*-mediated transformation, the RK2-derived *trfA* function for mobilization between *E. coli* and other hosts, and the *OriT* and *OriV* functions also from RK2. The pCIB2001 polylinker is suitable for the cloning of plant expression cassettes containing their own regulatory signals.

b. pCIB10 and Hygromycin Selection Derivatives thereof:

The binary vector pCIB10 contains a gene encoding kanamycin resistance for selection in plants and T-DNA right and left border sequences and incorporates sequences from the wide host-range plasmid pRK252 allowing it to replicate in both *E. coli* and *Agrobacterium*. Its construction is described by Rothstein *et al.* (1987). Various derivatives of pCIB10 are constructed which incorporate the gene for hygromycin B phosphotransferase described by Gritz *et al.*, 1983). These derivatives enable selection of transgenic plant cells on hygromycin only (pCIB743), or hygromycin and kanamycin (pCIB715, pCIB717).

2. Vectors Suitable for non-*Agrobacterium* Transformation

Transformation without the use of *Agrobacterium tumefaciens* circumvents the requirement for T-DNA sequences in the chosen transformation vector and consequently vectors lacking these sequences can be utilized in addition to vectors such as the ones described above which contain T-DNA sequences. Transformation techniques that do not rely on *Agrobacterium* include transformation via particle bombardment, protoplast uptake (*e.g.* PEG and electroporation) and microinjection. The choice of vector depends largely on the preferred selection for the species being transformed. Below, the construction of typical vectors suitable for non-*Agrobacterium* transformation is described.

a. pCIB3064:

pCIB3064 is a pUC-derived vector suitable for direct gene transfer techniques in combination with selection by the herbicide basta (or phosphinothricin). The plasmid pCIB246 comprises the CaMV 35S promoter in operational fusion to the *E. coli* GUS gene and the CaMV 35S transcriptional terminator and is described in the PCT published application WO 93/07278. The 35S promoter of this vector contains two ATG sequences 5' of the start site. These sites are mutated using standard PCR techniques in such a way as to remove the ATGs and generate the restriction sites *Sspl* and *PvuII*. The new restriction sites are 96 and 37 bp away from the unique *SalI* site and 101 and 42 bp away from the actual start site. The resultant derivative of pCIB246 is designated pCIB3025. The GUS gene is then excised from pCIB3025 by digestion with *SalI* and *SacI*, the termini rendered blunt and religated to generate plasmid pCIB3060. The plasmid pJIT82 is obtained from the John Innes Centre, Norwich and the a 400 bp *SmaI* fragment containing the *bar* gene from *Streptomyces viridochromogenes* is excised and inserted into the *HpaI* site of pCIB3060 (Thompson *et al.*, 1987). This generated pCIB3064, which comprises the *bar* gene under the control of the CaMV 35S promoter and terminator for herbicide selection, a gene for ampicillin resistance (for selection in *E. coli*) and a polylinker with the unique sites *SphI, PstI, HindIII,* and *BamHI*. This vector is suitable for the cloning of plant expression cassettes containing their own regulatory signals.

b. pSOG19 and pSOG35:

pSOG35 is a transformation vector that utilizes the *E. coli* gene dihydrofolate reductase (DFR) as a selectable marker conferring resistance to methotrexate. PCR is

used to amplify the 35S promoter (-800 bp), intron 6 from the maize Adh1 gene (-550 bp) and 18 bp of the GUS untranslated leader sequence from pSOG10. A 250-bp fragment encoding the *E. coli* dihydrofolate reductase type II gene is also amplified by PCR and these two PCR fragments are assembled with a *SacI-PstI* fragment from pB1221 (Clontech) which comprises the pUC19 vector backbone and the nopaline synthase terminator. Assembly of these fragments generates pSOG19 which contains the 35S promoter in fusion with the intron 6 sequence, the GUS leader, the DHFR gene and the nopaline synthase terminator. Replacement of the GUS leader in pSOG19 with the leader sequence from Maize Chlorotic Mottle Virus (MCMV) generates the vector pSOG35. pSOG19 and pSOG35 carry the pUC gene for ampicillin resistance and have *HindIII, SphI, PstI* and *EcoRI* sites available for the cloning of foreign substances.

Example 12: Transformation

Once the gene sequence of interest has been cloned into an expression system, it is transformed into a plant cell. Methods for transformation and regeneration of plants are well known in the art. For example, Ti plasmid vectors have been utilized for the delivery of foreign DNA, as well as direct DNA uptake, liposomes, electroporation, micro-injection, and microprojectiles. In addition, bacteria from the genus *Agrobacterium* can be utilized to transform plant cells. Below are descriptions of representative techniques for transforming both dicotyledonous and monocotyledonous plants.

1. Transformation of Dicotyledons

Transformation techniques for dicotyledons are well known in the art and include *Agrobacterium*-based techniques and techniques that do not require *Agrobacterium*. Non-*Agrobacterium* techniques involve the uptake of exogenous genetic material directly by protoplasts or cells. This can be accomplished by PEG or electroporation mediated uptake, particle bombardment-mediated delivery, or microinjection. Examples of these techniques are described by Paszkowski *et al.*, 1984; Potrykus *et al.*, 1985; Reich *et al.*, 1986; and Klein *et al.*, 1987. In each case the transformed cells are regenerated to whole plants using standard techniques known in the art.

Agrobacterium-mediated transformation is a preferred technique for transformation of dicotyledons because of its high efficiency of transformation and its broad utility with many different species. *Agrobacterium* transformation typically involves the transfer of the binary

vector carrying the foreign DNA of interest (*e.g.* pCIB200 or pCIB2001) to an appropriate *Agrobacterium* strain which may depend of the complement of *vir* genes carried by the host *Agrobacterium* strain either on a co-resident Ti plasmid or chromosomally (*e.g.* strain CIB542 for pCIB200 and pCIB2001 (Uknes *et al.*, 1993). The transfer of the recombinant binary vector to *Agrobacterium* is accomplished by a triparental mating procedure using *E. coli* carrying the recombinant binary vector, a helper *E. coli* strain which carries a plasmid such as pRK2013 and which is able to mobilize the recombinant binary vector to the target *Agrobacterium* strain. Alternatively, the recombinant binary vector can be transferred to *Agrobacterium* by DNA transformation (Höfgen & Willmitzer, 1988).

Transformation of the target plant species by recombinant *Agrobacterium* usually involves co-cultivation of the *Agrobacterium* with explants from the plant and follows protocols well known in the art. Transformed tissue is regenerated on selectable medium carrying the antibiotic or herbicide resistance marker present between the binary plasmid T-DNA borders.

Another approach to transforming plant cells with a gene involves propelling inert or biologically active particles at plant tissues and cells. This technique is disclosed in U.S. Patent Nos. 4,945,050, 5,036,006, and 5,100,792. Generally, this procedure involves propelling inert or biologically active particles at the cells under conditions effective to penetrate the outer surface of the cell and afford incorporation within the interior thereof. When inert particles are utilized, the vector can be introduced into the cell by coating the particles with the vector containing the desired gene. Alternatively, the target cell can be surrounded by the vector so that the vector is carried into the cell by the wake of the particle. Biologically active particles (e.g., dried yeast cells, dried bacterium or a bacteriophage, each containing DNA sought to be introduced) can also be propelled into plant cell tissue.

2. Transformation of Monocotyledons

Transformation of most monocotyledon species has now also become routine. Preferred techniques include direct gene transfer into protoplasts using PEG or electroporation techniques, and particle bombardment into callus tissue. Transformations can be undertaken with a single DNA species or multiple DNA species (*i.e.* co-transformation) and both these techniques are suitable for use with this invention. Co-transformation may have the advantage of avoiding complete vector construction and of generating transgenic plants with unlinked loci for the gene of interest and the selectable

marker, enabling the removal of the selectable marker in subsequent generations, should this be regarded desirable. However, a disadvantage of the use of co-transformation is the less than 100% frequency with which separate DNA species are integrated into the genome (Schocher *et al.*, 1986).

Patent Applications EP 0 292 435, EP 0 392 225, and WO 93/07278 describe techniques for the preparation of callus and protoplasts from an elite inbred line of maize, transformation of protoplasts using PEG or electroporation, and the regeneration of maize plants from transformed protoplasts. Gordon-Kamm *et al.* (1990) and Fromm *et al.* (1990) have published techniques for transformation of A188-derived maize line using particle bombardment. Furthermore, WO 93/07278 and Koziel *et al.* (1993) describe techniques for the transformation of elite inbred lines of maize by particle bombardment. This technique utilizes immature maize embryos of 1.5-2.5 mm length excised from a maize ear 14-15 days after pollination and a PDS-1000He Biolistics device for bombardment.

Transformation of rice can also be undertaken by direct gene transfer techniques utilizing protoplasts or particle bombardment. Protoplast-mediated transformation has been described for *Japonica*-types and *Indica*-types (Zhang *et al.*, 1988; Shimamoto *et al.*, 1989; Datta *et al.*, 1990). Both types are also routinely transformable using particle bombardment (Christou *et al.*, 1991). Furthermore, WO 93/21335 describes techniques for the transformation of rice via electroporation.

Patent Application EP 0 332 581 describes techniques for the generation, transformation and regeneration of Pooideae protoplasts. These techniques allow the transformation of *Dactylis* and wheat. Furthermore, wheat transformation has been described by Vasil *et al.* (1992) using particle bombardment into cells of type C long-term regenerable callus, and also by Vasil *et al.* (1993) and Weeks *et al.* (1993) using particle bombardment of immature embryos and immature embryo-derived callus. A preferred technique for wheat transformation, however, involves the transformation of wheat by particle bombardment of immature embryos and includes either a high sucrose or a high maltose step prior to gene delivery. Prior to bombardment, any number of embryos (0.75-1 mm in length) are plated onto MS medium with 3% sucrose (Murashiga & Skoog, 1962) and 3 mg/l 2,4-D for induction of somatic embryos, which is allowed to proceed in the dark. On the chosen day of bombardment, embryos are removed from the induction medium and placed onto the osmoticum (*i.e.* induction medium with sucrose or maltose added at the desired concentration, typically 15%). The embryos are allowed to plasmolyze for 2-3 h and are then bombarded. Twenty embryos per target plate is typical, although not critical. An

appropriate gene-carrying plasmid (such as pCIB3064 or pSG35) is precipitated onto micrometer size gold particles using standard procedures. Each plate of embryos is shot with the DuPont Biolistics® helium device using a burst pressure of ~1000 psi using a standard 80 mesh screen. After bombardment, the embryos are placed back into the dark to recover for about 24 h (still on osmoticum). After 24 hrs, the embryos are removed from the osmoticum and placed back onto induction medium where they stay for about a month before regeneration. Approximately one month later the embryo explants with developing embryogenic callus are transferred to regeneration medium (MS + 1 mg/liter NAA, 5 mg/liter GA), further containing the appropriate selection agent (10 mg/l basta in the case of pCIB3064 and 2 mg/l methotrexate in the case of pSOG35). After approximately one month, developed shoots are transferred to larger sterile containers known as "GA7s" which contain half-strength MS, 2% sucrose, and the same concentration of selection agent.

Tranformation of monocotyledons using *Agrobacterium* has also been described. *See,* WO 94/00977 and U.S. Patent No. 5,591,616.

III. Breeding and Seed Production

Example 13: Breeding

The plants obtained via tranformation with a gene of the present invention can be any of a wide variety of plant species, including those of monocots and dicots; however, the plants used in the method of the invention are preferably selected from the list of agronomically important target crops set forth *supra*. The expression of a gene of the present invention in combination with other characteristics important for production and quality can be incorporated into plant lines through breeding. Breeding approaches and techniques are known in the art. See, for example, Welsh J. R. (1981); Wood D. R. (Ed.) (1983); Mayo O. (1987); Singh, D.P. (1986); and Wricke and Weber (1986).

The genetic properties engineered into the transgenic seeds and plants described above are passed on by sexual reproduction or vegetative growth and can thus be maintained and propagated in progeny plants. Generally said maintenance and propagation make use of known agricultural methods developed to fit specific purposes such as tilling, sowing or harvesting. Specialized processes such as hydroponics or greenhouse technologies can also be applied. As the growing crop is vulnerable to attack and damages caused by insects or infections as well as to competition by weed plants, measures are

undertaken to control weeds, plant diseases, insects, nematodes, and other adverse conditions to improve yield. These include mechanical measures such a tillage of the soil or removal of weeds and infected plants, as well as the application of agrochemicals such as herbicides, fungicides, gametocides, nematicides, growth regulants, ripening agents and insecticides.

Use of the advantageous genetic properties of the transgenic plants and seeds according to the invention can further be made in plant breeding, which aims at the development of plants with improved properties such as tolerance of pests, herbicides, or stress, improved nutritional value, increased yield, or improved structure causing less loss from lodging or shattering. The various breeding steps are characterized by well-defined human intervention such as selecting the lines to be crossed, directing pollination of the parental lines, or selecting appropriate progeny plants. Depending on the desired properties, different breeding measures are taken. The relevant techniques are well known in the art and include but are not limited to hybridization, inbreeding, backcross breeding, multiline breeding, variety blend, interspecific hybridization, aneuploid techniques, etc. Hybridization techniques also include the sterilization of plants to yield male or female sterile plants by mechanical, chemical, or biochemical means. Cross pollination of a male sterile plant with pollen of a different line assures that the genome of the male sterile but female fertile plant will uniformly obtain properties of both parental lines. Thus, the transgenic seeds and plants according to the invention can be used for the breeding of improved plant lines, that for example, increase the effectiveness of conventional methods such as herbicide or pestidice treatment or allow one to dispense with said methods due to their modified genetic properties. Alternatively new crops with improved stress tolerance can be obtained, which, due to their optimized genetic "equipment", yield harvested product of better quality than products that were not able to tolerate comparable adverse developmental conditions.

Example 14: Seed Production

In seeds production, germination quality and uniformity of seeds are essential product characteristics, whereas germination quality and uniformity of seeds harvested and sold by the farmer is not important. As it is difficult to keep a crop free from other crop and weed seeds, to control seedborne diseases, and to produce seed with good germination, fairly extensive and well-defined seed production practices have been developed by seed

producers, who are experienced in the art of growing, conditioning and marketing of pure seed. Thus, it is common practice for the farmer to buy certified seed meeting specific quality standards instead of using seed harvested from his own crop. Propagation material to be used as seeds is customarily treated with a protectant coating comprising herbicides, insecticides, fungicides, bactericides, nematicides, molluscicides, or mixtures thereof. Customarily used protectant coatings comprise compounds such as captan, carboxin, thiram (TMTD®), methalaxyl (Apron®), and pirimiphos-methyl (Actellic®). If desired, these compounds are formulated together with further carriers, surfactants or application-promoting adjuvants customarily employed in the art of formulation to provide protection against damage caused by bacterial, fungal or animal pests. The protectant coatings may be applied by impregnating propagation material with a liquid formulation or by coating with a combined wet or dry formulation. Other methods of application are also possible such as treatment directed at the buds or the fruit.

It is a further aspect of the present invention to provide new agricultural methods, such as the methods examplified above, which are characterized by the use of transgenic plants, transgenic plant material, or transgenic seed according to the present invention.

The seeds may be provided in a bag, container or vessel comprised of a suitable packaging material, the bag or container capable of being closed to contain seeds. The bag, container or vessel may be designed for either short term or long term storage, or both, of the seed. Examples of a suitable packaging material include paper, such as kraft paper, rigid or pliable plastic or other polymeric material, glass or metal. Desirably the bag, container, or vessel is comprised of a plurality of layers of packaging materials, of the same or differing type. In one embodiment the bag, container or vessel is provided so as to exclude or limit water and moisture from contacting the seed. In one example, the bag, container or vessel is sealed, for example heat sealed, to prevent water or moisture from entering. In another embodiment water absorbent materials are placed between or adjacent to packaging material layers. In yet another embodiment the bag, container or vessel, or packaging material of which it is comprised is treated to limit, suppress or prevent disease, contamination or other adverse affects of storage or transport of the seed. An example of such treatment is sterilization, for example by chemical means or by exposure to radiation. Comprised by the present invention is a commercial bag comprising seed of a transgenic plant comprising a gene of the present invention that is expressed in said transformed plant at higher levels than in a wild type plant, together with a suitable

carrier, together with label instructions for the use thereof for conferring broad spectrum disease resistance to plants.

IV. Disease Resistance Evaluation

Disease resistance evaluation is performed by methods known in the art. *See*, Uknes *et al.* (1993); Görlach *et al.* (1996); Alexander *et al.* (1993). For example, several representative disease resistance assays are described below.

Example 15: *Phytophthora parasitica* (Black Shank) Resistance Assay

Assays for resistance to *Phytophthora parasitica*, the causative organism of black shank, are performed on six-week-old plants grown as described in Alexander *et al.* (1993). Plants are watered, allowed to drain well, and then inoculated by applying 10 ml of a sporangium suspension (300 sporangia/ml) to the soil. Inoculated plants are kept in a greenhouse maintained at 23-25°C day temperature, and 20-22°C night temperature. The wilt index used for the assay is as follows: 0=no symptoms; 1=no symptoms; 1=some sign of wilting, with reduced turgidity; 2=clear wilting symptoms, but no rotting or stunting; 3=clear wilting symptoms with stunting, but no apparent stem rot; 4=severe wilting, with visible stem rot and some damage to root system; 5=as for 4, but plants near death or dead, and with severe reduction of root system. All assays are scored blind on plants arrayed in a random design.

Example 16: *Pseudomonas syringae* Resistance Assay

Pseudomonas syringae pv. tabaci strain #551 is injected into the two lower leaves of several 6-7-week-old plants at a concentration of 10^6 or 3 x 10^6 per ml in H_20. Six individual plants are evaluated at each time point. *Pseudomonas tabaci* infected plants are rated on a 5 point disease severity scale, 5=100% dead tissue, 0=no symptoms. A T-test (LSD) is conducted on the evaluations for each day and the groupings are indicated after the Mean disease rating value. Values followed by the same letter on that day of evaluation are not statistically significantly different.

Example 17: *Cercospora nicotianae* Resistance Assay

A spore suspension of *Cercospora nicotianae* (ATCC #18366) (100,000-150,000 spores per ml) is sprayed to imminent run-off onto the surface of the leaves. The plants are maintained in 100% humidity for five days. Thereafter the plants are misted with water 5-10 times per day. Six individual plants are evaluated at each time point. *Cercospora nicotianae* is rated on a % leaf area showing disease symptoms basis. A T-test (LSD) is conducted on the evaluations for each day and the groupings are indicated after the Mean disease rating value. Values followed by the same letter on that day of evaluation are not statistically significantly different.

Example 18: *Peronospora parasitica* Resistance Assay

Assays for resistance to *Peronospora parasitica* are performed on plants as described in Uknes *et al*, (1993). Plants are inoculated with a compatible isolate of *P. parasitica* by spraying with a conidial suspension (approximately 5 x 10^4 spores per milliliter). Inoculated plants are incubated under humid conditions at 17° C in a growth chamber with a 14-hr day/10-hr night cycle. Plants are examined at 3-14 days, preferably 7-12 days, after inoculation for the presence of conidiophores. In addition, several plants from each treatment are randomly selected and stained with lactophenol-trypan blue (Keogh *et al.*, 1980) for microscopic examination.

The above disclosed embodiments are illustrative. This disclosure of the invention will place one skilled in the art in possession of many variations of the invention. All such obvious and foreseeable variations are intended to be encompassed by the claims.

BRIEF DESCRIPTION OF THE SEQUENCES IN THE SEQUENCE LISTING

SEQ ID NO:1 - Full length cDNA sequence of a *NIM1* homologue from *Nicotiana tabacum.*

SEQ ID NO:2 - Protein sequence of the *Nicotiana tabacum* NIM1 homologue encoded by SEQ ID NO:1.

SEQ ID NO:3 - Full length cDNA sequence of a *NIM1* homologue from *Lycopersicon esculentum.*

SEQ ID NO:4 - Protein sequence of the *Lycopersicon esculentum* NIM1 homologue encoded by SEQ ID NO:3.

SEQ ID NO:5 - Partial cDNA sequence of a *NIM1* homologue from *Brassica napus*.

SEQ ID NO:6 - Partial protein sequence of the *Brassica napus* NIM1 homologue encoded by SEQ ID NO:5.

SEQ ID NO:7 - Full length cDNA sequence of a *NIM1* homologue (*AtNMLc5*) from *Arabidopsis thaliana*.

SEQ ID NO:8 - Full length protein sequence of the *Arabidopsis thaliana* NIM1 homologue *AtNMLc5* encoded by SEQ ID NO:7.

SEQ ID NOs:9-14 - Oligonucleotide primers used in Examples 1-4.

SEQ ID NO:15 - Genomic DNA sequence of a *NIM1* homologue (*AtNMLc2*) from *Arabidopsis thaliana*.

SEQ ID NO:16 - Protein sequence of the *Arabidopsis thaliana* NIM1 homologue *AtNMLc2* encoded by SEQ ID NO:15.

SEQ ID NO:17 - Genomic DNA sequence of a *NIM1* homologue (*AtNMLc4-1*) from *Arabidopsis thaliana*.

SEQ ID NO:18 - Protein sequence of the *Arabidopsis thaliana* NIM1 homologue *AtNMLc4-1* encoded by SEQ ID NO:17.

SEQ ID NO:19 - Genomic DNA sequence of a *NIM1* homologue (*AtNMLc4-2*) from *Arabidopsis thaliana*.

SEQ ID NO:20 - Protein sequence of the *Arabidopsis thaliana* NIM1 homologue *AtNMLc4-2* encoded by SEQ ID NO:19.

SEQ ID NO:21 - PCR primer NIM 1A.

SEQ ID NO:22 - PCR primer NIM 1B.

SEQ ID NO:23 - PCR primer NIM 1C.

SEQ ID NO:24 - PCR primer NIM 1D.

SEQ ID NO:25 - PCR primer NIM 2A.

SEQ ID NO:26 - PCR primer NIM 2B.

SEQ ID NO:27 - PCR primer NIM 2C.

SEQ ID NO:28 - PCR primer NIM 2D.

SEQ ID NO:29 - 659 bp *NIM*-like DNA fragment amplified from *Nicotiana tabacum* (Tobacco A), which is a consensus of 36 sequences and has 67% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:30 - Protein sequence encoded by SEQ ID NO:29.

SEQ ID NO:31 - 498 bp *NIM*-like DNA fragment amplified from *Nicotiana tabacum* (Tobacco B), which is a consensus of 2 sequences and has 62% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:32 - Protein sequence encoded by SEQ ID NO:31.

SEQ ID NO:33 - 498 bp *NIM*-like DNA fragment amplified from *Nicotiana tabacum* (Tobacco C), which is a consensus of 3 sequences and has 63% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:34 - Protein sequence encoded by SEQ ID NO:33.

SEQ ID NO:35 - 399 bp *NIM*-like DNA fragment amplified from *Nicotiana tabacum* (Tobacco D), which has 59% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:36 - Protein sequence encoded by SEQ ID NO:35.

SEQ ID NO:37 - 498 bp *NIM*-like DNA fragment amplified from *Lycopersicon esculentum* (Tomato A), which is a consensus of 8 sequences and has 67% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:38 - Protein sequence encoded by SEQ ID NO:37.

SEQ ID NO:39 - 498 bp *NIM*-like DNA fragment amplified from *Beta vulgaris* (Sugarbeet), which is a consensus of 24 sequences and has 66% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:40 - Protein sequence encoded by SEQ ID NO:39.

SEQ ID NO:41 - 498 bp *NIM*-like DNA fragment amplified from *Helianthus annuus* (Sunflower A), which is a consensus of 9 sequences and has 61% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:42 - Protein sequence encoded by SEQ ID NO:41.

SEQ ID NO:43 - 498 bp *NIM*-like DNA fragment amplified from *Helianthus annuus* (Sunflower B), which is a consensus of 10 sequences and has 59% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:44 - Protein sequence encoded by SEQ ID NO:43.

SEQ ID NO:45 - 653 bp *NIM*-like DNA fragment amplified from *Solanum tuberosum* (Potato A), which is a consensus of 15 sequences and has 68% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:46 - Protein sequence encoded by SEQ ID NO:45.

SEQ ID NO:47 - 498 bp *NIM*-like DNA fragment amplified from *Solanum tuberosum* (Potato B), which is a consensus of 3 sequences and has 61% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:48 - Protein sequence encoded by SEQ ID NO:47.

SEQ ID NO:49 - 477 bp *NIM*-like DNA fragment amplified from *Solanum tuberosum* (Potato C), which is a consensus of 2 sequences and has 62% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:50 - Protein sequence encoded by SEQ ID NO:49.

SEQ ID NO:51 - 501 bp *NIM*-like DNA fragment amplified from *Brassica napus* (Canola A), which is a consensus of 5 sequences and has 59% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:52 - Protein sequence encoded by SEQ ID NO:51.

SEQ ID NO:53 - 501 bp *NIM*-like DNA fragment amplified from *Brassica napus* (Canola B), which is a consensus of 5 sequences and has 58% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:54 - Protein sequence encoded by SEQ ID NO:53.

SEQ ID NO:55 - 498 bp *NIM*-like DNA fragment amplified from *Brassica napus* (Canola C), which has 56% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:56 - Protein sequence encoded by SEQ ID NO:55.

SEQ ID NO:57 - 498 bp *NIM*-like DNA fragment amplified from *Brassica napus* (Canola D), which has 73% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:58 - Protein sequence encoded by SEQ ID NO:57.

SEQ ID NO:59 - PCR primer NIM 3A.

SEQ ID NO:60 - PCR primer NIM 3B.

SEQ ID NO:61 - 148 bp *NIM*-like DNA fragment amplified from *Lycopersicon esculentum* (Tomato B), which is a consensus of 3 sequences and has 72% sequence identity to the *Arabidopsis thaliana NIM1* gene sequence.

SEQ ID NO:62 - Protein sequence encoded by SEQ ID NO:61.

SEQ ID NO:63 - Full length cDNA sequence of a *NIM1* homologue from *Beta vulgaris* (Sugarbeet), which corresponds to the PCR fragment of SEQ ID NO:39.

SEQ ID NO:64 - Protein sequence of the sugarbeet NIM1 homologue encoded by SEQ ID NO:62.

SEQ ID NO:65 - Full length cDNA sequence of a *NIM1* homologue from *Helianthus annuus* (Sunflower B), which corresponds to the PCR fragment of SEQ ID NO:43.

SEQ ID NO:66 - Protein sequence of the *Helianthus annuus* NIM1 homologue encoded by SEQ ID NO:65.

SEQ ID NO:67 - cDNA sequence corresponding to the *Arabidopsis thaliana NIM*-like genomic sequence *AtNMLc2* (SEQ ID NO:15).

SEQ ID NO:68 - Protein sequence encoded by SEQ ID NO:67.

SEQ ID NO:69 - cDNA sequence corresponding to the *Arabidopsis thaliana NIM*-like genomic sequence *AtNMLc4-1* (SEQ ID NO:17).

SEQ ID NO:70 - Protein sequence encoded by SEQ ID NO:69.

SEQ ID NO:71 - cDNA sequence corresponding to the *Arabidopsis thaliana NIM*-like genomic sequence *AtNMLc4-2* (SEQ ID NO:19).

SEQ ID NO:72 - Protein sequence encoded by SEQ ID NO:71.

SEQ ID NO:73 - Full length cDNA sequence of a *NIM1* homologue from *Nicotiana tabacum* (Tobacco B), which corresponds to the PCR fragment of SEQ ID NO:31.

SEQ ID NO:74 - Protein sequence of the *Nicotiana tabacum* NIM1 homologue encoded by SEQ ID NO:73.

DEPOSITS

The following material has been deposited with the Agricultural Research Service, Patent Culture Collection (NRRL), 1815 North University Street, Peoria, Illinois 61604, USA, under the terms of the Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purposes of Patent Procedure. All restrictions on the availability of the deposited material will be irrevocably removed upon the granting of a patent.

Clone	Accession Number	Date of Deposit
pNOV1203	NRRL B-30049	August 17, 1998
pNOV1204	NRRL B-30050	August 17, 1998
pNOV1206	NRRL B-30051	August 17, 1998
AtNMLc5	NRRL B-30139	May 25, 1999

REFERENCES

The references cited herein are indicative of the current state of the art. Each of the following is incorporated by reference into the instant disclosure.

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U.S. Patent No. 4,945,050
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U.S. Patent No. 5,523,311
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INDICATIONS RELATING TO DEPOSITED MICROORGANISM OR OTHER BIOLOGICAL MATERIAL

(PCT Rule 13*bis*)

A. The indications made below relate to the deposited microorganism or other biological material referred to in the description on page 50, line 19-31

B. IDENTIFICATION OF DEPOSIT — Further deposits are identified on an additional sheet ☐

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Agricultural Research Service Culture Collection (NRRL)

Address of depositary institution *(including postal code and country)*
1815 North University Street
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United States of America (USA)

Date of deposit	Accession Number
17 August 1998 (17.08.98)	NRRL B-30049

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Date of deposit	Accession Number
17 August 1998 (17.08.98)	NRRL B-30051

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Address of depositary institution *(including postal code and country)*
1815 North University Street
Peoria, Illinois 61604
United States of America (USA)

Date of deposit	Accession Number
25 May 1999 (25.05.99)	NRRL B-30139

C. ADDITIONAL INDICATIONS *(leave blank if not applicable)* — This information is continued on an additional sheet ☐

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Claims

What Is Claimed Is:

1. An isolated nucleic acid molecule comprising:

(a) a nucleotide sequence that encodes SEQ ID NO:2, 4, 6, 8, 16, 18, 20, 30, 32, 34, 36, 38, 40, 42, 44, 46, 48, 50, 52, 54, 56, 58, 62, 64, 66, 68, 70, 72, or 74;

(b) SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73;

(c) a nucleotide sequence that comprises an at least 20 consecutive base pair portion identical in sequence to an at least 20 consecutive base pair portion of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73;

(d) a nucleotide sequence that can be amplified from a *Lycopersicon esculentum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, SEQ ID NO:26 and 28, or SEQ ID NO:59 and 60;

(e) a nucleotide sequence that can be amplified from a *Beta vulgaris* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:22 and 24 or SEQ ID NO:26 and 28;

(f) a nucleotide sequence that can be amplified from a *Helianthus annuus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:26 and 28;

(g) a nucleotide sequence that can be amplified from a *Solanum tuberosum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:21 and 24, SEQ ID NO:21 and 23, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28;

(h) a nucleotide sequence that can be amplified from a *Brassica napus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10 or SEQ ID NO:26 and 28;

(i) a nucleotide sequence that can be amplified from an *Arabidopsis thaliana* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:13 and 14, SEQ ID NO:21 and 24, or SEQ ID NO:22 and 24;

(j) a nucleotide sequence that can be amplified from an *Nicotiana tabacum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID

NO:9 and 10, SEQ ID NO:11 and 12, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28; or

(k) a nucleotide sequence that can be amplified from an plant DNA library using the polymerase chain reaction with a pair of primers comprising the first 20 nucleotides and the reverse complement of the last 20 nucleotides of the coding sequence (CDS) of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

2. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that encodes SEQ ID NO:2, 4, 6, 8, 16, 18, 20, 30, 32, 34, 36, 38, 40, 42, 44, 46, 48, 50, 52, 54, 56, 58, 62, 64, 66, 68, 70, 72, or 74.

3. An isolated nucleic acid molecule according to claim 1, comprising SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

4. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that comprises an at least 20 consecutive base pair portion identical in sequence to an at least 20 consecutive base pair portion of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

5. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a *Lycopersicon esculentum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, SEQ ID NO:26 and 28, or SEQ ID NO:59 and 60.

6. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a *Beta vulgaris* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:22 and 24 or SEQ ID NO:26 and 28.

7. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a *Helianthus annuus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:26 and 28.

8. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a *Solanum tuberosum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:21 and 24, SEQ ID NO:21 and 23, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28.

9. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a *Brassica napus* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10 or SEQ ID NO:26 and 28.

10. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from an *Arabidopsis thaliana* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:13 and 14, SEQ ID NO:21 and 24, or SEQ ID NO:22 and 24.

11. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from an *Nicotiana tabacum* DNA library using the polymerase chain reaction with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:11 and 12, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:25 and 28, or SEQ ID NO:26 and 28.

12. An isolated nucleic acid molecule according to claim 1, comprising a nucleotide sequence that can be amplified from a plant DNA library using the polymerase chain reaction with a pair of primers corresponding to the first 20 nucleotides and the reverse complement of the last 20 nucleotides of the coding sequence (CDS) of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73.

13. A chimeric gene comprising a promoter active in plants operatively linked to a nucleic acid molecule according to any one of the preceding claims.

14. A recombinant vector comprising the chimeric gene of claim 13.

15. A host cell comprising the chimeric gene of claim 13.

16. A plant comprising the chimeric gene of claim 13.

17. The plant of claim 16, which is selected from the following: rice, wheat, barley, rye, corn, potato, canola, sunflower, carrot, sweet potato, sugarbeet, bean, pea, chicory, lettuce, cabbage, cauliflower, broccoli, turnip, radish, spinach, asparagus, onion, garlic, eggplant, pepper, celery, squash, pumpkin, cucumber, apple, pear, quince, melon, plum, cherry, peach, nectarine, apricot, strawberry, grape, raspberry, blackberry, pineapple, avocado, papaya, mango, banana, soybean, tobacco, tomato, sorghum and sugarcane.

18. Seed from the plant of claim 16.

19. A method of increasing SAR gene expression in a plant, comprising expressing the chimeric gene of claim 13 in said plant.

20. A method of enhancing disease resistance in a plant, comprising expressing the chimeric gene of claim 13 in said plant.

21. A PCR primer selected from the group consisting of SEQ ID NO:9-14, 21-28, 59, and 60.

22. A method for isolating a *NIM1* homologue involved in the signal transduction cascade leading to systemic acquired resistance in plants comprising amplifying a DNA molecule from a plant DNA library using the polymerase chain reaction with a pair of primers corresponding to the first 20 nucleotides and the reverse complement of the last 20 nucleotides of the coding sequence (CDS) of SEQ ID NO:1, 3, 5, 7, 15, 17, 19, 29, 31, 33, 35, 37, 39, 41, 43, 45, 47, 49, 51, 53, 55, 57, 61, 63, 65, 67, 69, 71, or 73 or with the pair of primers set forth as SEQ ID NO:9 and 10, SEQ ID NO:11 and 12, SEQ ID NO:13 and 14, SEQ ID NO:21 and 24, SEQ ID NO:22 and 24, SEQ ID NO:21 and 23, SEQ ID NO:25 and 28, SEQ ID NO:26 and 28, or SEQ ID NO:59 and 60.

23. The method of claim 22, wherein said plant DNA library is a *Nicotiana tabacum* (tobacco), *Lycopersicon esculentum* (tomato), *Brassica napus* (oilseed rape), *Arabidopsis thaliana*, *Beta vulgaris* (sugarbeet), *Helianthus annuus* (sunflower), or *Solanum tuberosum* (potato) DNA library.

SEQUENCE LISTING

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<223> Full length tobacco cDNA sequence

<400> 1

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agc agt agt ata tgc tgc atc ggc ggc ggc atg act gaa ttt ttc tcg   96
Ser Ser Ser Ile Cys Cys Ile Gly Gly Gly Met Thr Glu Phe Phe Ser
             20                  25                  30

ccg gag act tcg ccg gcg gag atc act tca ctg aaa cgc cta tcg gaa   144
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         35                  40                  45

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gcc gac gct aag ctt gtg gtt tcc ggc ccg tgt aag gaa att ccg gtg   240
Ala Asp Ala Lys Leu Val Val Ser Gly Pro Cys Lys Glu Ile Pro Val
 65                  70                  75                  80

cac cgg tgc att ttg tcg gcg agg agt ccg ttc ttt aag aat ttg ttc   288
His Arg Cys Ile Leu Ser Ala Arg Ser Pro Phe Phe Lys Asn Leu Phe
                 85                  90                  95

tgc ggt aaa aag gag aag aat agt agt aag gtg gaa ttg aag gag gtg   336
Cys Gly Lys Lys Glu Lys Asn Ser Ser Lys Val Glu Leu Lys Glu Val
            100                 105                 110

atg aaa gag cat gag gtg agc tat gat gct gta atg agt gta ttg gct   384
Met Lys Glu His Glu Val Ser Tyr Asp Ala Val Met Ser Val Leu Ala
        115                 120                 125

tat ttg tat agt ggt aaa gtt agg cct tca cct aaa gat gtg tgt gtt   432
Tyr Leu Tyr Ser Gly Lys Val Arg Pro Ser Pro Lys Asp Val Cys Val
    130                 135                 140
```

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tgt gtg gac aat gac tgc tct cat gtg gct tgt agg cca gct gtg gca   480
Cys Val Asp Asn Asp Cys Ser His Val Ala Cys Arg Pro Ala Val Ala
145                 150                 155                 160

ttc ctg gtt gag gtt ttg tac aca tca ttt acc ttt cag atc tct gaa   528
Phe Leu Val Glu Val Leu Tyr Thr Ser Phe Thr Phe Gln Ile Ser Glu
                165                 170                 175

ttg gtt gac aag ttt cag aga cac cta ctg gat att ctt gac aaa act   576
Leu Val Asp Lys Phe Gln Arg His Leu Leu Asp Ile Leu Asp Lys Thr
            180                 185                 190

gca gca gac gat gta atg atg gtt tta tct gtt gca aac att tgt ggt   624
Ala Ala Asp Asp Val Met Met Val Leu Ser Val Ala Asn Ile Cys Gly
        195                 200                 205

aaa gca tgc gag aga ttg ctt tca agc tgc att gag att att gtc aag   672
Lys Ala Cys Glu Arg Leu Leu Ser Ser Cys Ile Glu Ile Ile Val Lys
    210                 215                 220

tct aat gtt gat atc ata acc ctt gat aaa gcc ttg cct cat gac att   720
Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ala Leu Pro His Asp Ile
225                 230                 235                 240

gta aaa caa att act gat tca cga gcg gaa ctt ggt cta caa ggg cct   768
Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu Gly Leu Gln Gly Pro
                245                 250                 255

gaa agc aac ggt ttt cct gat aaa cat gtt aag agg ata cat agg gca   816
Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys Arg Ile His Arg Ala
            260                 265                 270

ttg gat tct gat gat gtt gaa tta cta caa atg ttg cta aga gag ggg   864
Leu Asp Ser Asp Asp Val Glu Leu Leu Gln Met Leu Leu Arg Glu Gly
        275                 280                 285

cat act acc cta gat gat gca tat gct ctc cat tat gct gta gcg tat   912
His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala Tyr
    290                 295                 300

tgc gat gca aag act aca gca gaa ctt cta gat ctt gca ctt gct gat   960
Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp Leu Ala Leu Ala Asp
305                 310                 315                 320

att aat cat caa aat tca agg gga tac acg gtg ctg cat gtt gca gcc  1008
Ile Asn His Gln Asn Ser Arg Gly Tyr Thr Val Leu His Val Ala Ala
                325                 330                 335

atg agg aaa gag cct aaa att gta gtg tcc ctt tta acc aaa gga gct  1056
Met Arg Lys Glu Pro Lys Ile Val Val Ser Leu Leu Thr Lys Gly Ala
            340                 345                 350

aga cct tct gat ctg aca tcc gat gga aga aaa gca ctt caa atc gcc  1104
Arg Pro Ser Asp Leu Thr Ser Asp Gly Arg Lys Ala Leu Gln Ile Ala
        355                 360                 365

aag agg ctc act agg ctt gtg gat ttc agt aag tct ccg gag gaa gga  1152
Lys Arg Leu Thr Arg Leu Val Asp Phe Ser Lys Ser Pro Glu Glu Gly
    370                 375                 380
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aaa tct gct tcg aat gat cgg tta tgc att gag att ctg gag caa gca   1200
Lys Ser Ala Ser Asn Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln Ala
385                 390                 395                 400

gaa aga aga gac cct ctg cta gga gaa gct tct gta tct ctt gct atg   1248
Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser Val Ser Leu Ala Met
                405                 410                 415

gca ggc gat gat ttg cgt atg aag ctg tta tac ctt gaa aat aga gtt   1296
Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg Val
            420                 425                 430

ggc ctg gct aaa ctc ctt ttt cca atg gaa gct aaa gtt gca atg gac   1344
Gly Leu Ala Lys Leu Leu Phe Pro Met Glu Ala Lys Val Ala Met Asp
        435                 440                 445

att gct caa gtt gat ggc act tct gag ttc cca ctg gct agc atc ggc   1392
Ile Ala Gln Val Asp Gly Thr Ser Glu Phe Pro Leu Ala Ser Ile Gly
    450                 455                 460

aaa aag atg gct aat gca cag agg aca aca gta gat ttg aac gag gct   1440
Lys Lys Met Ala Asn Ala Gln Arg Thr Thr Val Asp Leu Asn Glu Ala
465                 470                 475                 480

cct ttc aag ata aaa gag gag cac ttg aat cgg ctt aga gca ctc tct   1488
Pro Phe Lys Ile Lys Glu Glu His Leu Asn Arg Leu Arg Ala Leu Ser
                485                 490                 495

aga act gta gaa ctt gga aaa cgc ttc ttt cca cgt tgt tca gaa gtt   1536
Arg Thr Val Glu Leu Gly Lys Arg Phe Phe Pro Arg Cys Ser Glu Val
            500                 505                 510

cta aat aag atc atg gat gct gat gac ttg tct gag ata gct tac atg   1584
Leu Asn Lys Ile Met Asp Ala Asp Asp Leu Ser Glu Ile Ala Tyr Met
        515                 520                 525

ggg aat gat acg gca gaa gag cgt caa ctg aag aag caa agg tac atg   1632
Gly Asn Asp Thr Ala Glu Glu Arg Gln Leu Lys Lys Gln Arg Tyr Met
    530                 535                 540

gaa ctt caa gaa att ctg act aaa gca ttc act gag gat aaa gaa gaa   1680
Glu Leu Gln Glu Ile Leu Thr Lys Ala Phe Thr Glu Asp Lys Glu Glu
545                 550                 555                 560

tat gat aag act aac aac atc tcc tca tct tgt tcc tct aca tct aag   1728
Tyr Asp Lys Thr Asn Asn Ile Ser Ser Ser Cys Ser Ser Thr Ser Lys
                565                 570                 575

gga gta gat aag ccc aat aag ctc cct ttt agg aaa tag               1767
Gly Val Asp Lys Pro Asn Lys Leu Pro Phe Arg Lys
            580                 585
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<210> 2
<211> 588
<212> PRT
<213> Nicotiana tabacum

<400> 2

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Met Asp Asn Ser Arg Thr Ala Phe Ser Asp Ser Asn Asp Ile Ser Gly
  1               5                  10                  15
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Ser Ser Ser Ile Cys Cys Ile Gly Gly Gly Met Thr Glu Phe Phe Ser
            20                  25                  30

Pro Glu Thr Ser Pro Ala Glu Ile Thr Ser Leu Lys Arg Leu Ser Glu
        35                  40                  45

Thr Leu Glu Ser Ile Phe Asp Ala Ser Leu Pro Glu Phe Asp Tyr Phe
    50                  55                  60

Ala Asp Ala Lys Leu Val Val Ser Gly Pro Cys Lys Glu Ile Pro Val
65                  70                  75                  80

His Arg Cys Ile Leu Ser Ala Arg Ser Pro Phe Phe Lys Asn Leu Phe
                85                  90                  95

Cys Gly Lys Lys Glu Lys Asn Ser Ser Lys Val Glu Leu Lys Glu Val
            100                 105                 110

Met Lys Glu His Glu Val Ser Tyr Asp Ala Val Met Ser Val Leu Ala
        115                 120                 125

Tyr Leu Tyr Ser Gly Lys Val Arg Pro Ser Pro Lys Asp Val Cys Val
    130                 135                 140

Cys Val Asp Asn Asp Cys Ser His Val Ala Cys Arg Pro Ala Val Ala
145                 150                 155                 160

Phe Leu Val Glu Val Leu Tyr Thr Ser Phe Thr Phe Gln Ile Ser Glu
                165                 170                 175

Leu Val Asp Lys Phe Gln Arg His Leu Leu Asp Ile Leu Asp Lys Thr
            180                 185                 190

Ala Ala Asp Asp Val Met Met Val Leu Ser Val Ala Asn Ile Cys Gly
        195                 200                 205

Lys Ala Cys Glu Arg Leu Leu Ser Ser Cys Ile Glu Ile Ile Val Lys
    210                 215                 220

Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ala Leu Pro His Asp Ile
225                 230                 235                 240

Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu Gly Leu Gln Gly Pro
                245                 250                 255

Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys Arg Ile His Arg Ala
            260                 265                 270

Leu Asp Ser Asp Asp Val Glu Leu Leu Gln Met Leu Leu Arg Glu Gly
        275                 280                 285

His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala Tyr
    290                 295                 300

Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp Leu Ala Leu Ala Asp
305                 310                 315                 320

Ile Asn His Gln Asn Ser Arg Gly Tyr Thr Val Leu His Val Ala Ala
                325                 330                 335
```

```
Met Arg Lys Glu Pro Lys Ile Val Val Ser Leu Leu Thr Lys Gly Ala
            340                 345                 350

Arg Pro Ser Asp Leu Thr Ser Asp Gly Arg Lys Ala Leu Gln Ile Ala
        355                 360                 365

Lys Arg Leu Thr Arg Leu Val Asp Phe Ser Lys Ser Pro Glu Glu Gly
    370                 375                 380

Lys Ser Ala Ser Asn Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln Ala
385                 390                 395                 400

Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser Val Ser Leu Ala Met
                405                 410                 415

Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg Val
            420                 425                 430

Gly Leu Ala Lys Leu Leu Phe Pro Met Glu Ala Lys Val Ala Met Asp
        435                 440                 445

Ile Ala Gln Val Asp Gly Thr Ser Glu Phe Pro Leu Ala Ser Ile Gly
    450                 455                 460

Lys Lys Met Ala Asn Ala Gln Arg Thr Thr Val Asp Leu Asn Glu Ala
465                 470                 475                 480

Pro Phe Lys Ile Lys Glu Glu His Leu Asn Arg Leu Arg Ala Leu Ser
                485                 490                 495

Arg Thr Val Glu Leu Gly Lys Arg Phe Phe Pro Arg Cys Ser Glu Val
            500                 505                 510

Leu Asn Lys Ile Met Asp Ala Asp Asp Leu Ser Glu Ile Ala Tyr Met
        515                 520                 525

Gly Asn Asp Thr Ala Glu Glu Arg Gln Leu Lys Lys Gln Arg Tyr Met
    530                 535                 540

Glu Leu Gln Glu Ile Leu Thr Lys Ala Phe Thr Glu Asp Lys Glu Glu
545                 550                 555                 560

Tyr Asp Lys Thr Asn Asn Ile Ser Ser Ser Cys Ser Ser Thr Ser Lys
                565                 570                 575

Gly Val Asp Lys Pro Asn Lys Leu Pro Phe Arg Lys
            580                 585
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<210> 3
<211> 1731
<212> DNA
<213> Lycopersicon esculentum

<220>
<221> CDS
<222> (1)..(1728)
<223> Full length tomato cDNA sequence

<400> 3

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atg gat agt aga act gct ttt tcg gat tcc aat gat att agt gga agc   48
Met Asp Ser Arg Thr Ala Phe Ser Asp Ser Asn Asp Ile Ser Gly Ser
  1               5                  10                  15

agt agt ata tgc tgc atg aac gaa tcg gaa act tca ctg gca gac gtc   96
Ser Ser Ile Cys Cys Met Asn Glu Ser Glu Thr Ser Leu Ala Asp Val
            20                  25                  30

aat tcc ctc aaa cgt cta tca gaa aca cta gag tct atc ttc gat gcg   144
Asn Ser Leu Lys Arg Leu Ser Glu Thr Leu Glu Ser Ile Phe Asp Ala
        35                  40                  45

tct gcg ccg gat ttc gac ttc ttc gct gat gct aag ctt ctg gct cca   192
Ser Ala Pro Asp Phe Asp Phe Phe Ala Asp Ala Lys Leu Leu Ala Pro
    50                  55                  60

ggc ggt aag gaa att ccg gtg cat cgg tgc att ttg tcg gcg agg agt   240
Gly Gly Lys Glu Ile Pro Val His Arg Cys Ile Leu Ser Ala Arg Ser
65                  70                  75                  80

cct ttt ttt aag aat gta ttc tgt ggg aaa gat agc agc acg aag ctg   288
Pro Phe Phe Lys Asn Val Phe Cys Gly Lys Asp Ser Ser Thr Lys Leu
                85                  90                  95

gaa ctc aaa gag ctg atg aaa gag tat gag gtg agt ttt gat gcc gtg   336
Glu Leu Lys Glu Leu Met Lys Glu Tyr Glu Val Ser Phe Asp Ala Val
            100                 105                 110

gtc agt gtg ctc gcc tat ttg tat agt gga aaa gtt agg cct gca tct   384
Val Ser Val Leu Ala Tyr Leu Tyr Ser Gly Lys Val Arg Pro Ala Ser
        115                 120                 125

aaa gat gtg tgt gtt tgt gtg gac aat gag tgc ttg cat gta gct tgt   432
Lys Asp Val Cys Val Cys Val Asp Asn Glu Cys Leu His Val Ala Cys
    130                 135                 140

agg cca gct gtg gcc ttc atg gtt cag gtt ttg tac gca tcc ttt acc   480
Arg Pro Ala Val Ala Phe Met Val Gln Val Leu Tyr Ala Ser Phe Thr
145                 150                 155                 160

ttt cag atc tct caa ttg gtc gac aag ttt cag aga cac cta ttg gat   528
Phe Gln Ile Ser Gln Leu Val Asp Lys Phe Gln Arg His Leu Leu Asp
                165                 170                 175

att ctt gac aaa gct gta gca gat gat gta atg atg gtt tta tcc gtt   576
Ile Leu Asp Lys Ala Val Ala Asp Asp Val Met Met Val Leu Ser Val
            180                 185                 190

gca aac att tgc ggt aaa gca tgt gaa aga tta ctt tca aga tgc att   624
Ala Asn Ile Cys Gly Lys Ala Cys Glu Arg Leu Leu Ser Arg Cys Ile
        195                 200                 205

gat att att gtc aag tct aat gtt gat atc ata acc ctt gat aag tcc   672
Asp Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ser
    210                 215                 220

ttg cct cat gac att gta aaa caa atc act gat tca cgt gct gaa ctt   720
Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu
225                 230                 235                 240

ggt ctg caa ggg cct gaa agc aat ggt ttt cct gat aaa cat gtt aag   768
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```
Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys
                245                 250                 255

agg ata cat aga gca ttg gac tct gat gat gtt gaa tta cta agg atg   816
Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met
            260                 265                 270

ttg ctt aaa gag ggg cat act act ctt gat gat gca tat gct ctc cac   864
Leu Leu Lys Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His
        275                 280                 285

tat gct gta gca tat tgc gat gca aag act aca gca gaa ctt tta gat   912
Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp
    290                 295                 300

ctt tca ctt gct gat gtt aat cat caa aat cct aga gga cac acg gta   960
Leu Ser Leu Ala Asp Val Asn His Gln Asn Pro Arg Gly His Thr Val
305                 310                 315                 320

ctt cat gtt gct gcc atg agg aaa gaa cct aaa att ata gtg tcc ctt  1008
Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu
                325                 330                 335

tta acc aaa gga gct aga cct tct gat ctg aca tcc gat ggc aaa aaa  1056
Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys
            340                 345                 350

gca ctt caa att gct aag agg ctc act agg ctt gta gat ttt acc aag  1104
Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys
        355                 360                 365

tct aca gag gaa gga aaa tct gct cca aag gat cgg tta tgc att gag  1152
Ser Thr Glu Glu Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu
    370                 375                 380

att ctg gag caa gca gaa aga aga gat cca cta cta gga gaa gct tca  1200
Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser
385                 390                 395                 400

tta tct ctt gct atg gca ggc gat gat ttg cgt atg aag ctg tta tac  1248
Leu Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr
                405                 410                 415

ctt gaa aat aga gtt ggt ctg gct aaa ctc ctt ttt ccc atg gaa gca  1296
Leu Glu Asn Arg Val Gly Leu Ala Lys Leu Leu Phe Pro Met Glu Ala
            420                 425                 430

aaa gtt gca atg gac att gca caa gtt gat ggc acg tct gaa tta ccc  1344
Lys Val Ala Met Asp Ile Ala Gln Val Asp Gly Thr Ser Glu Leu Pro
        435                 440                 445

ctg gct agc atg agg aag aag ata gct gat gca cag agg aca aca gtg  1392
Leu Ala Ser Met Arg Lys Lys Ile Ala Asp Ala Gln Arg Thr Thr Val
    450                 455                 460

gat ttg aac gag gct cct ttc aag atg aaa gag gag cac ttg aat cgg  1440
Asp Leu Asn Glu Ala Pro Phe Lys Met Lys Glu Glu His Leu Asn Arg
465                 470                 475                 480

ctt agg gct ctc tct aga act gtg gaa ctt gga aaa cgg ttc ttt cca  1488
Leu Arg Ala Leu Ser Arg Thr Val Glu Leu Gly Lys Arg Phe Phe Pro
```

```
                485                 490                 495

cgt tgt tca gaa gtt cta aat aag atc atg gat gct gat gac ttg tct   1536
Arg Cys Ser Glu Val Leu Asn Lys Ile Met Asp Ala Asp Asp Leu Ser
            500                 505                 510

gag ata gct tac atg ggg aat gat aca gta gaa gag cgt caa ctg aag   1584
Glu Ile Ala Tyr Met Gly Asn Asp Thr Val Glu Glu Arg Gln Leu Lys
        515                 520                 525

aag caa agg tac atg gaa ctt caa gaa att ttg tct aaa gca ttc acg   1632
Lys Gln Arg Tyr Met Glu Leu Gln Glu Ile Leu Ser Lys Ala Phe Thr
    530                 535                 540

gag gat aaa gaa gaa ttt gct aag act aac atg tcc tca tct tgt tcc   1680
Glu Asp Lys Glu Glu Phe Ala Lys Thr Asn Met Ser Ser Ser Cys Ser
545                 550                 555                 560

tct aca tct aag gga gta gat aag ccc aat aat ctc cca ttt agg aaa   1728
Ser Thr Ser Lys Gly Val Asp Lys Pro Asn Asn Leu Pro Phe Arg Lys
                565                 570                 575

tag                                                              1731
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<210> 4
<211> 576
<212> PRT
<213> Lycopersicon esculentum

<400> 4

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Met Asp Ser Arg Thr Ala Phe Ser Asp Ser Asn Asp Ile Ser Gly Ser
  1               5                  10                  15

Ser Ser Ile Cys Cys Met Asn Glu Ser Glu Thr Ser Leu Ala Asp Val
            20                  25                  30

Asn Ser Leu Lys Arg Leu Ser Glu Thr Leu Glu Ser Ile Phe Asp Ala
        35                  40                  45

Ser Ala Pro Asp Phe Asp Phe Phe Ala Asp Ala Lys Leu Leu Ala Pro
    50                  55                  60

Gly Gly Lys Glu Ile Pro Val His Arg Cys Ile Leu Ser Ala Arg Ser
65                  70                  75                  80

Pro Phe Phe Lys Asn Val Phe Cys Gly Lys Asp Ser Ser Thr Lys Leu
                85                  90                  95

Glu Leu Lys Glu Leu Met Lys Glu Tyr Glu Val Ser Phe Asp Ala Val
            100                 105                 110

Val Ser Val Leu Ala Tyr Leu Tyr Ser Gly Lys Val Arg Pro Ala Ser
        115                 120                 125

Lys Asp Val Cys Val Cys Val Asp Asn Glu Cys Leu His Val Ala Cys
    130                 135                 140

Arg Pro Ala Val Ala Phe Met Val Gln Val Leu Tyr Ala Ser Phe Thr
145                 150                 155                 160
```

```
Phe Gln Ile Ser Gln Leu Val Asp Lys Phe Gln Arg His Leu Leu Asp
                165                 170                 175

Ile Leu Asp Lys Ala Val Ala Asp Asp Val Met Met Val Leu Ser Val
            180                 185                 190

Ala Asn Ile Cys Gly Lys Ala Cys Glu Arg Leu Leu Ser Arg Cys Ile
        195                 200                 205

Asp Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ser
    210                 215                 220

Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu
225                 230                 235                 240

Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys
                245                 250                 255

Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met
            260                 265                 270

Leu Leu Lys Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His
        275                 280                 285

Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp
    290                 295                 300

Leu Ser Leu Ala Asp Val Asn His Gln Asn Pro Arg Gly His Thr Val
305                 310                 315                 320

Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu
                325                 330                 335

Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys
            340                 345                 350

Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys
        355                 360                 365

Ser Thr Glu Glu Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu
    370                 375                 380

Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser
385                 390                 395                 400

Leu Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr
                405                 410                 415

Leu Glu Asn Arg Val Gly Leu Ala Lys Leu Leu Phe Pro Met Glu Ala
            420                 425                 430

Lys Val Ala Met Asp Ile Ala Gln Val Asp Gly Thr Ser Glu Leu Pro
        435                 440                 445

Leu Ala Ser Met Arg Lys Lys Ile Ala Asp Ala Gln Arg Thr Thr Val
    450                 455                 460

Asp Leu Asn Glu Ala Pro Phe Lys Met Lys Glu Glu His Leu Asn Arg
465                 470                 475                 480

Leu Arg Ala Leu Ser Arg Thr Val Glu Leu Gly Lys Arg Phe Phe Pro
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```
                485                 490                 495

Arg Cys Ser Glu Val Leu Asn Lys Ile Met Asp Ala Asp Asp Leu Ser
            500                 505                 510

Glu Ile Ala Tyr Met Gly Asn Asp Thr Val Glu Glu Arg Gln Leu Lys
        515                 520                 525

Lys Gln Arg Tyr Met Glu Leu Gln Glu Ile Leu Ser Lys Ala Phe Thr
    530                 535                 540

Glu Asp Lys Glu Glu Phe Ala Lys Thr Asn Met Ser Ser Ser Cys Ser
545                 550                 555                 560

Ser Thr Ser Lys Gly Val Asp Lys Pro Asn Asn Leu Pro Phe Arg Lys
                565                 570                 575
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<210> 5
<211> 1740
<212> DNA
<213> Brassica napus

<220>
<221> CDS
<222> (1)..(1737)
<223> Canola cDNA sequence

<400> 5

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Met Glu Thr Ile Ala Xaa Phe Asp Asp Phe Tyr Glu Ile Ser Ser Thr
  1               5                  10                  15

agc ttc cyc gcc gca ccg gcg cca acc gat aac tcc gga tca tcc acc   96
Ser Phe Xaa Ala Ala Pro Ala Pro Thr Asp Asn Ser Gly Ser Ser Thr
            20                  25                  30

gtc twc ccg acg gag ctt ytc acc aga ccc gag gta tcc gcg ttt caa   144
Val Xaa Pro Thr Glu Leu Xaa Thr Arg Pro Glu Val Ser Ala Phe Gln
        35                  40                  45

ctc ctc tcc aac agc ctc gag tcc gtc ttc gac tcg ccg gaa gcg ttc   192
Leu Leu Ser Asn Ser Leu Glu Ser Val Phe Asp Ser Pro Glu Ala Phe
    50                  55                  60

tac agc gac gcc aag ctt gtt ctc tcc gac gac aag gaa gta tcc ttc   240
Tyr Ser Asp Ala Lys Leu Val Leu Ser Asp Asp Lys Glu Val Ser Phe
65                  70                  75                  80

cac cgt tgc att ctc tcg gcg aga agc ctc ttc ttc aag gcc gct ttg   288
His Arg Cys Ile Leu Ser Ala Arg Ser Leu Phe Phe Lys Ala Ala Leu
                85                  90                  95

rca gcc gcc gag aag gtg cag aag tcc acc ccc gtg aag ctc gag ctg   336
Xaa Ala Ala Glu Lys Val Gln Lys Ser Thr Pro Val Lys Leu Glu Leu
            100                 105                 110

aag aca ctc gcg gcg gaa tac gac gtc ggg ttc gat tct gtg gtg gct   384
Lys Thr Leu Ala Ala Glu Tyr Asp Val Gly Phe Asp Ser Val Val Ala
        115                 120                 125
```

```
gtt ctg gcg tac gtt tac agc ggc aga gtg agg ccg cct ccg aag gga   432
Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg Pro Pro Pro Lys Gly
    130                 135                 140

gtt tct gaa tgc gca gac gak agc tgc tgc cac gtg gcg tgc cgt ccg   480
Val Ser Glu Cys Ala Asp Xaa Ser Cys Cys His Val Ala Cys Arg Pro
145                 150                 155                 160

gct gtg gat ttc atg gtg gag gtt ctc tac ttg gct ttc gtc ttc cag   528
Ala Val Asp Phe Met Val Glu Val Leu Tyr Leu Ala Phe Val Phe Gln
                165                 170                 175

att cag gaa ctg gtt acc atg tat cag agg cat tta ctg gat gtt gta   576
Ile Gln Glu Leu Val Thr Met Tyr Gln Arg His Leu Leu Asp Val Val
            180                 185                 190

gac aaa gtt awc ata gaa gac act ttg gtc gtc ctc aag ctt gct aac   624
Asp Lys Val Xaa Ile Glu Asp Thr Leu Val Val Leu Lys Leu Ala Asn
        195                 200                 205

atc tgc ggt aaa gcg tgc aag aag cta ttc gat aag tgc aga gag atc   672
Ile Cys Gly Lys Ala Cys Lys Lys Leu Phe Asp Lys Cys Arg Glu Ile
    210                 215                 220

att gtc aag tct aac gtg gat gtt gtt act cta aag aag tca ttg cct   720
Ile Val Lys Ser Asn Val Asp Val Val Thr Leu Lys Lys Ser Leu Pro
225                 230                 235                 240

gag rac att gcc aag caa gta atc gat atc cgc aaa gag ctc ggc ttg   768
Glu Xaa Ile Ala Lys Gln Val Ile Asp Ile Arg Lys Glu Leu Gly Leu
                245                 250                 255

gag gta gct gaa cca gag aaa cat gtc tcc aac ata cac aag gcg ctt   816
Glu Val Ala Glu Pro Glu Lys His Val Ser Asn Ile His Lys Ala Leu
            260                 265                 270

gag tca gac gat ctt gac ctt gtc gtt atg ctt ttg aaa gag ggc cac   864
Glu Ser Asp Asp Leu Asp Leu Val Val Met Leu Leu Lys Glu Gly His
        275                 280                 285

acg aat cta gac gaa gcg tat gct ctc cat ttt gct gtt gcg tat tgc   912
Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe Ala Val Ala Tyr Cys
    290                 295                 300

gat gag aag aca gcg agg aat ctc ctg gaa ctg ggg ttt gcg gat gtc   960
Asp Glu Lys Thr Ala Arg Asn Leu Leu Glu Leu Gly Phe Ala Asp Val
305                 310                 315                 320

aac cgg aga aac ccg aga ggg tac acg gta att cac gtc gct gcg atg   1008
Asn Arg Arg Asn Pro Arg Gly Tyr Thr Val Ile His Val Ala Ala Met
                325                 330                 335

agg aaa gag ccg aca ctg ata gca ttg ttg ttg acg aaa ggg gct aat   1056
Arg Lys Glu Pro Thr Leu Ile Ala Leu Leu Leu Thr Lys Gly Ala Asn
            340                 345                 350

gca tta gaa atg tct ttg gac ggg aga act gct ctg ttg atc gcg aaa   1104
Ala Leu Glu Met Ser Leu Asp Gly Arg Thr Ala Leu Leu Ile Ala Lys
        355                 360                 365
```

```
caa gtc act aag gcg gcc gag tgt tgt att ctg gag aaa ggg aag tta   1152
Gln Val Thr Lys Ala Ala Glu Cys Cys Ile Leu Glu Lys Gly Lys Leu
    370                 375                 380

gct gcc aaa ggc gga gta tgt gta gag ata ctc aag caa cca gac aac   1200
Ala Ala Lys Gly Gly Val Cys Val Glu Ile Leu Lys Gln Pro Asp Asn
385                 390                 395                 400

aca cga gaa cca ttt cct gaa gat gtt tct ccc tcc ctt gca gtg gct   1248
Thr Arg Glu Pro Phe Pro Glu Asp Val Ser Pro Ser Leu Ala Val Ala
                405                 410                 415

gct gat caa ttc aag ata agg ttg att gat ctt gaa aac aga gtt caa   1296
Ala Asp Gln Phe Lys Ile Arg Leu Ile Asp Leu Glu Asn Arg Val Gln
            420                 425                 430

atg gct cga tgt ctc tat cca atg gaa gca caa gtt gca atg gat ttc   1344
Met Ala Arg Cys Leu Tyr Pro Met Glu Ala Gln Val Ala Met Asp Phe
        435                 440                 445

gcc cga atg aag gga aca cgc gag ttt gtc gtg acg aca gca act gac   1392
Ala Arg Met Lys Gly Thr Arg Glu Phe Val Val Thr Thr Ala Thr Asp
    450                 455                 460

cta cac atg gaa cct ttc aag ttc gta gaa atg cat cag agt aga cta   1440
Leu His Met Glu Pro Phe Lys Phe Val Glu Met His Gln Ser Arg Leu
465                 470                 475                 480

aca gcg ctt tct aaa act gtg gaa ttc ggg aaa cgc ttc ttc cca cgc   1488
Thr Ala Leu Ser Lys Thr Val Glu Phe Gly Lys Arg Phe Phe Pro Arg
                485                 490                 495

tgt tcg aaa gtg ctc gat gat att gtg gac tct gag gac ttg act ata   1536
Cys Ser Lys Val Leu Asp Asp Ile Val Asp Ser Glu Asp Leu Thr Ile
            500                 505                 510

ctg gct ctc gta gaa gaa gac act cct gag caa cga caa caa aag agg   1584
Leu Ala Leu Val Glu Glu Asp Thr Pro Glu Gln Arg Gln Gln Lys Arg
        515                 520                 525

cag agg ttc atg gaa ata cag gag att gtt caa atg gcg ttt agt aaa   1632
Gln Arg Phe Met Glu Ile Gln Glu Ile Val Gln Met Ala Phe Ser Lys
    530                 535                 540

gac aag gag gat ctt gga aag tcg tct ctc tca gct tcg tct tct tcc   1680
Asp Lys Glu Asp Leu Gly Lys Ser Ser Leu Ser Ala Ser Ser Ser Ser
545                 550                 555                 560

aca tcc aaa tta act ggt aaa aag agg tct att gct aaa ccc tct cac   1728
Thr Ser Lys Leu Thr Gly Lys Lys Arg Ser Ile Ala Lys Pro Ser His
                565                 570                 575

cgg cgt cgg tga                                                  1740
Arg Arg Arg
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<210> 6
<211> 579
<212> PRT
<213> Brassica napus

<400> 6

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Met Glu Thr Ile Ala Xaa Phe Asp Asp Phe Tyr Glu Ile Ser Ser Thr
 1               5                  10                  15

Ser Phe Xaa Ala Ala Pro Ala Pro Thr Asp Asn Ser Gly Ser Ser Thr
            20                  25                  30

Val Xaa Pro Thr Glu Leu Xaa Thr Arg Pro Glu Val Ser Ala Phe Gln
        35                  40                  45

Leu Leu Ser Asn Ser Leu Glu Ser Val Phe Asp Ser Pro Glu Ala Phe
    50                  55                  60

Tyr Ser Asp Ala Lys Leu Val Leu Ser Asp Asp Lys Glu Val Ser Phe
65                  70                  75                  80

His Arg Cys Ile Leu Ser Ala Arg Ser Leu Phe Phe Lys Ala Ala Leu
                85                  90                  95

Xaa Ala Ala Glu Lys Val Gln Lys Ser Thr Pro Val Lys Leu Glu Leu
            100                 105                 110

Lys Thr Leu Ala Ala Glu Tyr Asp Val Gly Phe Asp Ser Val Val Ala
        115                 120                 125

Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg Pro Pro Pro Lys Gly
    130                 135                 140

Val Ser Glu Cys Ala Asp Xaa Ser Cys Cys His Val Ala Cys Arg Pro
145                 150                 155                 160

Ala Val Asp Phe Met Val Glu Val Leu Tyr Leu Ala Phe Val Phe Gln
                165                 170                 175

Ile Gln Glu Leu Val Thr Met Tyr Gln Arg His Leu Leu Asp Val Val
            180                 185                 190

Asp Lys Val Xaa Ile Glu Asp Thr Leu Val Val Leu Lys Leu Ala Asn
        195                 200                 205

Ile Cys Gly Lys Ala Cys Lys Lys Leu Phe Asp Lys Cys Arg Glu Ile
    210                 215                 220

Ile Val Lys Ser Asn Val Asp Val Val Thr Leu Lys Lys Ser Leu Pro
225                 230                 235                 240

Glu Xaa Ile Ala Lys Gln Val Ile Asp Ile Arg Lys Glu Leu Gly Leu
                245                 250                 255

Glu Val Ala Glu Pro Glu Lys His Val Ser Asn Ile His Lys Ala Leu
            260                 265                 270

Glu Ser Asp Asp Leu Asp Leu Val Val Met Leu Leu Lys Glu Gly His
        275                 280                 285

Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe Ala Val Ala Tyr Cys
    290                 295                 300

Asp Glu Lys Thr Ala Arg Asn Leu Leu Glu Leu Gly Phe Ala Asp Val
305                 310                 315                 320
```

```
Asn Arg Arg Asn Pro Arg Gly Tyr Thr Val Ile His Val Ala Ala Met
                325                 330                 335

Arg Lys Glu Pro Thr Leu Ile Ala Leu Leu Leu Thr Lys Gly Ala Asn
            340                 345                 350

Ala Leu Glu Met Ser Leu Asp Gly Arg Thr Ala Leu Leu Ile Ala Lys
        355                 360                 365

Gln Val Thr Lys Ala Ala Glu Cys Cys Ile Leu Glu Lys Gly Lys Leu
    370                 375                 380

Ala Ala Lys Gly Gly Val Cys Val Glu Ile Leu Lys Gln Pro Asp Asn
385                 390                 395                 400

Thr Arg Glu Pro Phe Pro Glu Asp Val Ser Pro Ser Leu Ala Val Ala
                405                 410                 415

Ala Asp Gln Phe Lys Ile Arg Leu Ile Asp Leu Glu Asn Arg Val Gln
            420                 425                 430

Met Ala Arg Cys Leu Tyr Pro Met Glu Ala Gln Val Ala Met Asp Phe
        435                 440                 445

Ala Arg Met Lys Gly Thr Arg Glu Phe Val Val Thr Thr Ala Thr Asp
    450                 455                 460

Leu His Met Glu Pro Phe Lys Phe Val Glu Met His Gln Ser Arg Leu
465                 470                 475                 480

Thr Ala Leu Ser Lys Thr Val Glu Phe Gly Lys Arg Phe Phe Pro Arg
                485                 490                 495

Cys Ser Lys Val Leu Asp Asp Ile Val Asp Ser Glu Asp Leu Thr Ile
            500                 505                 510

Leu Ala Leu Val Glu Glu Asp Thr Pro Glu Gln Arg Gln Gln Lys Arg
        515                 520                 525

Gln Arg Phe Met Glu Ile Gln Glu Ile Val Gln Met Ala Phe Ser Lys
    530                 535                 540

Asp Lys Glu Asp Leu Gly Lys Ser Ser Leu Ser Ala Ser Ser Ser Ser
545                 550                 555                 560

Thr Ser Lys Leu Thr Gly Lys Lys Arg Ser Ile Ala Lys Pro Ser His
                565                 570                 575

Arg Arg Arg
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<210> 7
<211> 1761
<212> DNA
<213> Arabidopsis thaliana

<220>
<221> CDS
<222> (1)..(1758)

<223> AtNMLc5 cDNA sequence

<400> 7

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atg gct act ttg act gag cca tca tca tct ttg agt ttc aca tct tct   48
Met Ala Thr Leu Thr Glu Pro Ser Ser Ser Leu Ser Phe Thr Ser Ser
  1               5                  10                  15

cat ttc tct tat ggt tct att ggg tcc aat cac ttc tca tca agc tca   96
His Phe Ser Tyr Gly Ser Ile Gly Ser Asn His Phe Ser Ser Ser Ser
            20                  25                  30

gct tct aat cct gaa gtt gtt agt cta acc aaa ctc agc tcc aat ctt   144
Ala Ser Asn Pro Glu Val Val Ser Leu Thr Lys Leu Ser Ser Asn Leu
        35                  40                  45

gag cag ctt ctt agt aat tca gat tgt gat tac agt gat gca gag atc   192
Glu Gln Leu Leu Ser Asn Ser Asp Cys Asp Tyr Ser Asp Ala Glu Ile
    50                  55                  60

att gtt gat ggt gtt cca gtt ggt gtt cat aga tgc att tta gct gca   240
Ile Val Asp Gly Val Pro Val Gly Val His Arg Cys Ile Leu Ala Ala
65                  70                  75                  80

aga agt aag ttt ttc caa gat ttg ttt aag aaa gaa aag aaa att tcg   288
Arg Ser Lys Phe Phe Gln Asp Leu Phe Lys Lys Glu Lys Lys Ile Ser
                85                  90                  95

aaa act gag aaa cca aag tat cag ttg aga gag atg tta cct tat gga   336
Lys Thr Glu Lys Pro Lys Tyr Gln Leu Arg Glu Met Leu Pro Tyr Gly
            100                 105                 110

gct gtt gct cat gaa gct ttc ttg tat ttc ttg agt tat ata tat act   384
Ala Val Ala His Glu Ala Phe Leu Tyr Phe Leu Ser Tyr Ile Tyr Thr
        115                 120                 125

ggg aga tta aag cct ttt cca ttg gag gtt tcg act tgt gtt gat cca   432
Gly Arg Leu Lys Pro Phe Pro Leu Glu Val Ser Thr Cys Val Asp Pro
    130                 135                 140

gtt tgt tct cat gat tgt tgt cga cct gcc att gat ttt gtt gtt caa   480
Val Cys Ser His Asp Cys Cys Arg Pro Ala Ile Asp Phe Val Val Gln
145                 150                 155                 160

ttg atg tat gct tcc tct gtt ctc caa gtg cct gag cta gtt tca tct   528
Leu Met Tyr Ala Ser Ser Val Leu Gln Val Pro Glu Leu Val Ser Ser
                165                 170                 175

ttt cag cgg cgg ctt tgt aac ttt gtg gag aag acc ctt gtt gag aat   576
Phe Gln Arg Arg Leu Cys Asn Phe Val Glu Lys Thr Leu Val Glu Asn
            180                 185                 190

gtt ctt ccc att ctt atg gtt gct ttc aat tgt aag ttg act cag ctt   624
Val Leu Pro Ile Leu Met Val Ala Phe Asn Cys Lys Leu Thr Gln Leu
        195                 200                 205

ctt gat cag tgt att gag aga gtg gcg agg tca gat ctt tac agg ttc   672
Leu Asp Gln Cys Ile Glu Arg Val Ala Arg Ser Asp Leu Tyr Arg Phe
    210                 215                 220

tgt att gaa aag gaa gtt cct ccc gaa gta gca gag aag att aaa cag   720
Cys Ile Glu Lys Glu Val Pro Pro Glu Val Ala Glu Lys Ile Lys Gln
```

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225                 230                 235                 240

ctt cga ctt ata tcc ccg caa gac gaa gaa acc agt ccc aag att tcg   768
Leu Arg Leu Ile Ser Pro Gln Asp Glu Glu Thr Ser Pro Lys Ile Ser
                245                 250                 255

gag aaa ttg ctt gaa aga atc ggt aaa att ctc aag gcc ttg gat tca   816
Glu Lys Leu Leu Glu Arg Ile Gly Lys Ile Leu Lys Ala Leu Asp Ser
            260                 265                 270

gat gat gtt gag ctt gtg aag ctt ctt ttg act gag tca gat atc act   864
Asp Asp Val Glu Leu Val Lys Leu Leu Leu Thr Glu Ser Asp Ile Thr
        275                 280                 285

cta gat caa gcc aat ggt ctg cat tat tct gtt gtg tat agt gat ccg   912
Leu Asp Gln Ala Asn Gly Leu His Tyr Ser Val Val Tyr Ser Asp Pro
    290                 295                 300

aaa gtt gtt gcc gag att ctt gct ctg gat atg ggt gat gtg aac tac   960
Lys Val Val Ala Glu Ile Leu Ala Leu Asp Met Gly Asp Val Asn Tyr
305                 310                 315                 320

agg aat tcc cgg ggt tac acg gtt ctt cat ttt gct gcg atg cgt aga   1008
Arg Asn Ser Arg Gly Tyr Thr Val Leu His Phe Ala Ala Met Arg Arg
                325                 330                 335

gag cca tcg atc att ata tcg ctt atc gat aaa ggc gcc aat gca tct   1056
Glu Pro Ser Ile Ile Ile Ser Leu Ile Asp Lys Gly Ala Asn Ala Ser
            340                 345                 350

gag ttt aca tct gac gga cgc agc gca gtt aat ata ttg aga aga ctg   1104
Glu Phe Thr Ser Asp Gly Arg Ser Ala Val Asn Ile Leu Arg Arg Leu
        355                 360                 365

aca aat cca aag gat tat cat acc aaa aca gca aaa ggg cgt gaa tct   1152
Thr Asn Pro Lys Asp Tyr His Thr Lys Thr Ala Lys Gly Arg Glu Ser
    370                 375                 380

agt aag gcc agg cta tgc atc gat ata ttg gaa aga gaa atc agg aag   1200
Ser Lys Ala Arg Leu Cys Ile Asp Ile Leu Glu Arg Glu Ile Arg Lys
385                 390                 395                 400

aac ccc atg gtt cta gat aca cca atg tgt tcc att tct atg cct gaa   1248
Asn Pro Met Val Leu Asp Thr Pro Met Cys Ser Ile Ser Met Pro Glu
                405                 410                 415

gat ctc cag atg aga ctg ttg tac cta gaa aag aga gtg ggt ctt gct   1296
Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Lys Arg Val Gly Leu Ala
            420                 425                 430

cag ttg ttc ttt cca acg gaa gct aaa gtg gct atg gac att ggt aac   1344
Gln Leu Phe Phe Pro Thr Glu Ala Lys Val Ala Met Asp Ile Gly Asn
        435                 440                 445

gta gaa ggt aca agt gag ttc aca ggg ttg tca cct cct tca agt ggg   1392
Val Glu Gly Thr Ser Glu Phe Thr Gly Leu Ser Pro Pro Ser Ser Gly
    450                 455                 460

tta acc gga aac ttg agt cag gtt gat tta aac gaa act cct cat atg   1440
Leu Thr Gly Asn Leu Ser Gln Val Asp Leu Asn Glu Thr Pro His Met
465                 470                 475                 480
```

```
caa acc caa aga ctt ctt act cgt atg gtg gct cta atg aaa aca gtt   1488
Gln Thr Gln Arg Leu Leu Thr Arg Met Val Ala Leu Met Lys Thr Val
                485                 490                 495

gag act ggt cga agg ttt ttt cca tat ggt tca gag gtt cta gat aag   1536
Glu Thr Gly Arg Arg Phe Phe Pro Tyr Gly Ser Glu Val Leu Asp Lys
            500                 505                 510

tac atg gct gag tat ata gac gac gac atc ctc gac gat ttc cat ttt   1584
Tyr Met Ala Glu Tyr Ile Asp Asp Asp Ile Leu Asp Asp Phe His Phe
        515                 520                 525

gag aag gga tct aca cat gaa aga aga ttg aaa aga atg aga tat aga   1632
Glu Lys Gly Ser Thr His Glu Arg Arg Leu Lys Arg Met Arg Tyr Arg
    530                 535                 540

gag ctt aag gat gat gtc caa aag gca tat agc aaa gac aaa gag tct   1680
Glu Leu Lys Asp Asp Val Gln Lys Ala Tyr Ser Lys Asp Lys Glu Ser
545                 550                 555                 560

aag att gcg cgg tct tgt ctt tct gct tca tct tct cct tct tct tct   1728
Lys Ile Ala Arg Ser Cys Leu Ser Ala Ser Ser Ser Pro Ser Ser Ser
                565                 570                 575

tcc ata aga gat gat ctg cac aac aca aca tga                       1761
Ser Ile Arg Asp Asp Leu His Asn Thr Thr
            580                 585
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<210> 8
<211> 586
<212> PRT
<213> Arabidopsis thaliana

<400> 8

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Met Ala Thr Leu Thr Glu Pro Ser Ser Ser Leu Ser Phe Thr Ser Ser
  1               5                  10                  15

His Phe Ser Tyr Gly Ser Ile Gly Ser Asn His Phe Ser Ser Ser Ser
             20                  25                  30

Ala Ser Asn Pro Glu Val Val Ser Leu Thr Lys Leu Ser Ser Asn Leu
         35                  40                  45

Glu Gln Leu Leu Ser Asn Ser Asp Cys Asp Tyr Ser Asp Ala Glu Ile
     50                  55                  60

Ile Val Asp Gly Val Pro Val Gly Val His Arg Cys Ile Leu Ala Ala
 65                  70                  75                  80

Arg Ser Lys Phe Phe Gln Asp Leu Phe Lys Lys Glu Lys Lys Ile Ser
                 85                  90                  95

Lys Thr Glu Lys Pro Lys Tyr Gln Leu Arg Glu Met Leu Pro Tyr Gly
            100                 105                 110

Ala Val Ala His Glu Ala Phe Leu Tyr Phe Leu Ser Tyr Ile Tyr Thr
        115                 120                 125

Gly Arg Leu Lys Pro Phe Pro Leu Glu Val Ser Thr Cys Val Asp Pro
```

```
    130                 135                 140

Val Cys Ser His Asp Cys Cys Arg Pro Ala Ile Asp Phe Val Val Gln
145                 150                 155                 160

Leu Met Tyr Ala Ser Ser Val Leu Gln Val Pro Glu Leu Val Ser Ser
                165                 170                 175

Phe Gln Arg Arg Leu Cys Asn Phe Val Glu Lys Thr Leu Val Glu Asn
            180                 185                 190

Val Leu Pro Ile Leu Met Val Ala Phe Asn Cys Lys Leu Thr Gln Leu
        195                 200                 205

Leu Asp Gln Cys Ile Glu Arg Val Ala Arg Ser Asp Leu Tyr Arg Phe
    210                 215                 220

Cys Ile Glu Lys Glu Val Pro Pro Glu Val Ala Glu Lys Ile Lys Gln
225                 230                 235                 240

Leu Arg Leu Ile Ser Pro Gln Asp Glu Glu Thr Ser Pro Lys Ile Ser
                245                 250                 255

Glu Lys Leu Leu Glu Arg Ile Gly Lys Ile Leu Lys Ala Leu Asp Ser
            260                 265                 270

Asp Asp Val Glu Leu Val Lys Leu Leu Leu Thr Glu Ser Asp Ile Thr
        275                 280                 285

Leu Asp Gln Ala Asn Gly Leu His Tyr Ser Val Val Tyr Ser Asp Pro
    290                 295                 300

Lys Val Val Ala Glu Ile Leu Ala Leu Asp Met Gly Asp Val Asn Tyr
305                 310                 315                 320

Arg Asn Ser Arg Gly Tyr Thr Val Leu His Phe Ala Ala Met Arg Arg
                325                 330                 335

Glu Pro Ser Ile Ile Ile Ser Leu Ile Asp Lys Gly Ala Asn Ala Ser
            340                 345                 350

Glu Phe Thr Ser Asp Gly Arg Ser Ala Val Asn Ile Leu Arg Arg Leu
        355                 360                 365

Thr Asn Pro Lys Asp Tyr His Thr Lys Thr Ala Lys Gly Arg Glu Ser
    370                 375                 380

Ser Lys Ala Arg Leu Cys Ile Asp Ile Leu Glu Arg Glu Ile Arg Lys
385                 390                 395                 400

Asn Pro Met Val Leu Asp Thr Pro Met Cys Ser Ile Ser Met Pro Glu
                405                 410                 415

Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Lys Arg Val Gly Leu Ala
            420                 425                 430

Gln Leu Phe Phe Pro Thr Glu Ala Lys Val Ala Met Asp Ile Gly Asn
        435                 440                 445

Val Glu Gly Thr Ser Glu Phe Thr Gly Leu Ser Pro Pro Ser Ser Gly
    450                 455                 460
```

```
Leu Thr Gly Asn Leu Ser Gln Val Asp Leu Asn Glu Thr Pro His Met
465                 470                 475                 480

Gln Thr Gln Arg Leu Leu Thr Arg Met Val Ala Leu Met Lys Thr Val
                485                 490                 495

Glu Thr Gly Arg Arg Phe Phe Pro Tyr Gly Ser Glu Val Leu Asp Lys
            500                 505                 510

Tyr Met Ala Glu Tyr Ile Asp Asp Asp Ile Leu Asp Asp Phe His Phe
        515                 520                 525

Glu Lys Gly Ser Thr His Glu Arg Arg Leu Lys Arg Met Arg Tyr Arg
    530                 535                 540

Glu Leu Lys Asp Asp Val Gln Lys Ala Tyr Ser Lys Asp Lys Glu Ser
545                 550                 555                 560

Lys Ile Ala Arg Ser Cys Leu Ser Ala Ser Ser Ser Pro Ser Ser Ser
                565                 570                 575

Ser Ile Arg Asp Asp Leu His Asn Thr Thr
            580                 585
```

<210> 9
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 9
agattattgt caagtctaat g 21

<210> 10
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 10
ttccatgtac ctttgcttc 19

<210> 11
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 11
gcggatccat ggataatagt agg 23

<210> 12
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 12
gcggatccta tttcctaaaa ggg 23

<210> 13
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 13
tcaaggcctt ggattcagat g 21

<210> 14
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR Primer

<400> 14
attaactgcg ctacgtccgt c 21

<210> 15
<211> 1477
<212> DNA
<213> Arabidopsis thaliana

<220>
<221> CDS
<222> (1)..(1476)
<223> AtNMLc2 genomic sequence

<400> 15
atg agc aat ctt gaa gaa tct ttg aga tct cta tcg ttg gat ttc ctg 48
Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
1 5 10 15

aac cta cta atc aac ggt caa gct ttc tcc gac gtg act ttc agc gtt 96
Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
20 25 30

gaa ggt cgt tta gtc cac gct cac cgt tgt atc ctc gcc gca cgg agt 144
Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Ser
35 40 45

```
ctt ttc ttc cgc aaa ttc ttt tgt ggg aca gac tca cca caa cct gtc   192
Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
     50                  55                  60

aca ggt ata gac ccg acc caa cat ggg tcc gta ccc gct agc cca aca   240
Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
 65                  70                  75                  80

aga ggc tcc acg gcc cca gct gga att ata cca gtg aac tca gtc ggt   288
Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val Asn Ser Val Gly
                 85                  90                  95

tat gag gtt ttt ctg ttg cta ctt cag ttt ctt tat agc gga caa gtc   336
Tyr Glu Val Phe Leu Leu Leu Leu Gln Phe Leu Tyr Ser Gly Gln Val
            100                 105                 110

tcc atc gtg ccg cag aaa cac gag cct aga cct aat tgt ggc gag aga   384
Ser Ile Val Pro Gln Lys His Glu Pro Arg Pro Asn Cys Gly Glu Arg
        115                 120                 125

gga tgt tgg cac act cat tgc tca gcc gcc gtt gat ctt gct ctt gat   432
Gly Cys Trp His Thr His Cys Ser Ala Ala Val Asp Leu Ala Leu Asp
    130                 135                 140

act ctc gcc gcc tct cgt tac ttc ggc gtc gag cag ctc gca ttg ctc   480
Thr Leu Ala Ala Ser Arg Tyr Phe Gly Val Glu Gln Leu Ala Leu Leu
145                 150                 155                 160

acc cag aaa caa ttg gca agc atg gtg gag aaa gcc tct atc gaa gat   528
Thr Gln Lys Gln Leu Ala Ser Met Val Glu Lys Ala Ser Ile Glu Asp
                165                 170                 175

gtg atg aaa gtt tta ata gca tca aga aag caa gac atg cat caa tta   576
Val Met Lys Val Leu Ile Ala Ser Arg Lys Gln Asp Met His Gln Leu
            180                 185                 190

tgg acc acc tgc tct cac tta gtt atg agc aat ctt gaa gaa tct ttg   624
Trp Thr Thr Cys Ser His Leu Val Met Ser Asn Leu Glu Glu Ser Leu
        195                 200                 205

aga tct cta tcg ttg gat ttc ctg aac cta cta atc aac ggt caa gct   672
Arg Ser Leu Ser Leu Asp Phe Leu Asn Leu Leu Ile Asn Gly Gln Ala
    210                 215                 220

ttc tcc gac gtg act ttc agc gtt gaa ggt cgt tta gtc cac gct cac   720
Phe Ser Asp Val Thr Phe Ser Val Glu Gly Arg Leu Val His Ala His
225                 230                 235                 240

cgt tgt atc ctc gcc gca cgg agt ctt ttc ttc cgc aaa ttc ttt tgt   768
Arg Cys Ile Leu Ala Ala Arg Ser Leu Phe Phe Arg Lys Phe Phe Cys
                245                 250                 255

ggg aca gac tca cca caa cct gtc aca ggt ata gac ccg acc caa cat   816
Gly Thr Asp Ser Pro Gln Pro Val Thr Gly Ile Asp Pro Thr Gln His
            260                 265                 270

ggg tcc gta ccc gct agc cca aca aga ggc tcc acg gcc cca gct gga   864
Gly Ser Val Pro Ala Ser Pro Thr Arg Gly Ser Thr Ala Pro Ala Gly
        275                 280                 285

att ata cca gtg aac tca gtc ggt tat gag gtt ttt ctg ttg cta ctt   912
```

```
Ile Ile Pro Val Asn Ser Val Gly Tyr Glu Val Phe Leu Leu Leu Leu
    290                 295                 300

cag ttt ctt tat agc gga caa gtc tcc atc gtg ccg cag aaa cac gag   960
Gln Phe Leu Tyr Ser Gly Gln Val Ser Ile Val Pro Gln Lys His Glu
305                 310                 315                 320

cct aga cct aat tgt ggc gag aga gga tgt tgg cac act cat tgc tca   1008
Pro Arg Pro Asn Cys Gly Glu Arg Gly Cys Trp His Thr His Cys Ser
                325                 330                 335

gcc gcc gtt gat ctt gct ctt gat act ctc gcc gcc tct cgt tac ttc   1056
Ala Ala Val Asp Leu Ala Leu Asp Thr Leu Ala Ala Ser Arg Tyr Phe
            340                 345                 350

ggc gtc gag cag ctc gca ttg ctc acc cag aaa caa ttg gca agc atg   1104
Gly Val Glu Gln Leu Ala Leu Leu Thr Gln Lys Gln Leu Ala Ser Met
        355                 360                 365

gtg gag aaa gcc tct atc gaa gat gtg atg aaa gtt tta ata gca tca   1152
Val Glu Lys Ala Ser Ile Glu Asp Val Met Lys Val Leu Ile Ala Ser
    370                 375                 380

aga aag caa gac atg cat caa tta tgg acc acc tgc tct cac tta gtt   1200
Arg Lys Gln Asp Met His Gln Leu Trp Thr Thr Cys Ser His Leu Val
385                 390                 395                 400

atg agc aat ctt gaa gaa tct ttg aga tct cta tcg ttg gat ttc ctg   1248
Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
                405                 410                 415

aac cta cta atc aac ggt caa gct ttc tcc gac gtg act ttc agc gtt   1296
Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
            420                 425                 430

gaa ggt cgt tta gtc cac gct cac cgt tgt atc ctc gcc gca cgg agt   1344
Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Ser
        435                 440                 445

ctt ttc ttc cgc aaa ttc ttt tgt ggg aca gac tca cca caa cct gtc   1392
Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
    450                 455                 460

aca ggt ata gac ccg acc caa cat ggg tcc gta ccc gct agc cca aca   1440
Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
465                 470                 475                 480

aga ggc tcc acg gcc cca gct gga att ata cca gtg a                 1477
Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val
                485                 490


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<213> Arabidopsis thaliana

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Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
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Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
            20                  25                  30

Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Ser
        35                  40                  45

Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
    50                  55                  60

Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
65                  70                  75                  80

Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val Asn Ser Val Gly
                85                  90                  95

Tyr Glu Val Phe Leu Leu Leu Leu Gln Phe Leu Tyr Ser Gly Gln Val
            100                 105                 110

Ser Ile Val Pro Gln Lys His Glu Pro Arg Pro Asn Cys Gly Glu Arg
        115                 120                 125

Gly Cys Trp His Thr His Cys Ser Ala Ala Val Asp Leu Ala Leu Asp
    130                 135                 140

Thr Leu Ala Ala Ser Arg Tyr Phe Gly Val Glu Gln Leu Ala Leu Leu
145                 150                 155                 160

Thr Gln Lys Gln Leu Ala Ser Met Val Glu Lys Ala Ser Ile Glu Asp
                165                 170                 175

Val Met Lys Val Leu Ile Ala Ser Arg Lys Gln Asp Met His Gln Leu
            180                 185                 190

Trp Thr Thr Cys Ser His Leu Val Met Ser Asn Leu Glu Glu Ser Leu
        195                 200                 205

Arg Ser Leu Ser Leu Asp Phe Leu Asn Leu Leu Ile Asn Gly Gln Ala
    210                 215                 220

Phe Ser Asp Val Thr Phe Ser Val Glu Gly Arg Leu Val His Ala His
225                 230                 235                 240

Arg Cys Ile Leu Ala Ala Arg Ser Leu Phe Phe Arg Lys Phe Phe Cys
                245                 250                 255

Gly Thr Asp Ser Pro Gln Pro Val Thr Gly Ile Asp Pro Thr Gln His
            260                 265                 270

Gly Ser Val Pro Ala Ser Pro Thr Arg Gly Ser Thr Ala Pro Ala Gly
        275                 280                 285

Ile Ile Pro Val Asn Ser Val Gly Tyr Glu Val Phe Leu Leu Leu Leu
    290                 295                 300

Gln Phe Leu Tyr Ser Gly Gln Val Ser Ile Val Pro Gln Lys His Glu
305                 310                 315                 320

Pro Arg Pro Asn Cys Gly Glu Arg Gly Cys Trp His Thr His Cys Ser
                325                 330                 335

Ala Ala Val Asp Leu Ala Leu Asp Thr Leu Ala Ala Ser Arg Tyr Phe
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            340                 345                 350

Gly Val Glu Gln Leu Ala Leu Leu Thr Gln Lys Gln Leu Ala Ser Met
        355                 360                 365

Val Glu Lys Ala Ser Ile Glu Asp Val Met Lys Val Leu Ile Ala Ser
    370                 375                 380

Arg Lys Gln Asp Met His Gln Leu Trp Thr Thr Cys Ser His Leu Val
385                 390                 395                 400

Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
                405                 410                 415

Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
            420                 425                 430

Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Ser
        435                 440                 445

Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
    450                 455                 460

Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
465                 470                 475                 480

Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val
                485                 490
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<210> 17
<211> 1804
<212> DNA
<213> Arabidopsis thaliana

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<222> (1)..(1803)
<223> AtNMLc4-1 genomic sequence

<400> 17

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Met Ala Ala Thr Ala Ile Glu Pro Ser Ser Ser Ile Ser Phe Thr Ser
  1               5                  10                  15

tct cac tta tca aac cct tct cct gtt gtt act act tat cac tca gct   96
Ser His Leu Ser Asn Pro Ser Pro Val Val Thr Thr Tyr His Ser Ala
             20                  25                  30

gct aat ctt gaa gag ctc agc tct aac ttg gag cag ctt ctc act aat   144
Ala Asn Leu Glu Glu Leu Ser Ser Asn Leu Glu Gln Leu Leu Thr Asn
         35                  40                  45

cca gat tgc gat tac act gac gca gag atc atc att gaa gaa gaa gct   192
Pro Asp Cys Asp Tyr Thr Asp Ala Glu Ile Ile Ile Glu Glu Glu Ala
     50                  55                  60

aac cct gtg agt gtt cat aga tgt gtt tta gct gct agg agc aag ttt   240
Asn Pro Val Ser Val His Arg Cys Val Leu Ala Ala Arg Ser Lys Phe
 65                  70                  75                  80
```

```
ttt ctt gat ctg ttt aag aaa gat aaa gat agt agt gag aag aaa cct   288
Phe Leu Asp Leu Phe Lys Lys Asp Lys Asp Ser Ser Glu Lys Lys Pro
                 85                  90                  95

aag tat caa atg aaa gat tta tta cca tat gga aat gtg gga cgt gag   336
Lys Tyr Gln Met Lys Asp Leu Leu Pro Tyr Gly Asn Val Gly Arg Glu
            100                 105                 110

gca ttt ctg cat ttc ttg agc tat atc tac act ggg agg tta aag cct   384
Ala Phe Leu His Phe Leu Ser Tyr Ile Tyr Thr Gly Arg Leu Lys Pro
        115                 120                 125

ttt cct atc gag gtt tca act tgt gtt gat tca gtt tgt gct cat gat   432
Phe Pro Ile Glu Val Ser Thr Cys Val Asp Ser Val Cys Ala His Asp
    130                 135                 140

tct tgt aaa ccg gcc att gat ttt gct gtt gag ttg atg tat gct tca   480
Ser Cys Lys Pro Ala Ile Asp Phe Ala Val Glu Leu Met Tyr Ala Ser
145                 150                 155                 160

ttt gtg ttc caa atc ccg gat ctt gtt tcg tca ttt cag cgg aag ctt   528
Phe Val Phe Gln Ile Pro Asp Leu Val Ser Ser Phe Gln Arg Lys Leu
                165                 170                 175

cgt aac tat gtt gag aag tca cta gta gag aat gtt ctt cct atc ctc   576
Arg Asn Tyr Val Glu Lys Ser Leu Val Glu Asn Val Leu Pro Ile Leu
            180                 185                 190

tta gtt gcg ttt cat tgt gat ttg aca cag ctt ctt gat caa tgc att   624
Leu Val Ala Phe His Cys Asp Leu Thr Gln Leu Leu Asp Gln Cys Ile
        195                 200                 205

gag aga gtg gcg aga tca gac tta gac aga ttc tgt atc gaa aag gag   672
Glu Arg Val Ala Arg Ser Asp Leu Asp Arg Phe Cys Ile Glu Lys Glu
    210                 215                 220

ctt cct tta gaa gta ttg gaa aaa atc aaa cag ctt cga gtt aag tcg   720
Leu Pro Leu Glu Val Leu Glu Lys Ile Lys Gln Leu Arg Val Lys Ser
225                 230                 235                 240

gtg aac ata ccc gag gtg gag gat aaa tcg ata gag aga aca ggg aaa   768
Val Asn Ile Pro Glu Val Glu Asp Lys Ser Ile Glu Arg Thr Gly Lys
                245                 250                 255

gta ctc aag gca ttg gat tca gat gat gta gaa ctc gtg aag ctt ctt   816
Val Leu Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
            260                 265                 270

ttg act gag tca gat ata act cta gac caa gcc aat ggt cta cat tat   864
Leu Thr Glu Ser Asp Ile Thr Leu Asp Gln Ala Asn Gly Leu His Tyr
        275                 280                 285

gca gtg gca tac agt gat ccg aaa gtt gtg aca cag gtt ctt gat cta   912
Ala Val Ala Tyr Ser Asp Pro Lys Val Val Thr Gln Val Leu Asp Leu
    290                 295                 300

gat atg gct gat gtt aat ttc aga aat tcc agg ggg tat acg gtt ctt   960
Asp Met Ala Asp Val Asn Phe Arg Asn Ser Arg Gly Tyr Thr Val Leu
305                 310                 315                 320
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cat att gct gct atg cgt aga gag cca aca att atc ata cca ctt att   1008
His Ile Ala Ala Met Arg Arg Glu Pro Thr Ile Ile Ile Pro Leu Ile
                325                 330                 335

caa aaa gga gct aat gct tca gat ttc acg ttt gat gga cgc agt gcg   1056
Gln Lys Gly Ala Asn Ala Ser Asp Phe Thr Phe Asp Gly Arg Ser Ala
            340                 345                 350

gta aat ata tgt agg aga ctc act agg ccg aaa gat tat cat acc aaa   1104
Val Asn Ile Cys Arg Arg Leu Thr Arg Pro Lys Asp Tyr His Thr Lys
        355                 360                 365

acc tca agg aaa gaa cct agt aaa tac cgc tta tgc atc gat atc ttg   1152
Thr Ser Arg Lys Glu Pro Ser Lys Tyr Arg Leu Cys Ile Asp Ile Leu
    370                 375                 380

gaa agg gaa att aga agg aat cca ttg gtt agt ggg gat aca ccc act   1200
Glu Arg Glu Ile Arg Arg Asn Pro Leu Val Ser Gly Asp Thr Pro Thr
385                 390                 395                 400

tgt tcc cat tcg atg ccc gag gat ctc caa atg agg ttg tta tac tta   1248
Cys Ser His Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu
                405                 410                 415

gaa aag cga tgg gac ttg cgt cag ttg ttc ttc cca gca gaa gcc aat   1296
Glu Lys Arg Trp Asp Leu Arg Gln Leu Phe Phe Pro Ala Glu Ala Asn
            420                 425                 430

gtg gct atg gac gtt gct aat gtt gaa ggg aca agc gag tgc aca ggt   1344
Val Ala Met Asp Val Ala Asn Val Glu Gly Thr Ser Glu Cys Thr Gly
        435                 440                 445

ctt cta act cca cct cca tca aat gat aca act gaa aac ttg ggt aaa   1392
Leu Leu Thr Pro Pro Pro Ser Asn Asp Thr Thr Glu Asn Leu Gly Lys
    450                 455                 460

gtc gat tta aat gaa acg cct tat gtg caa acg aaa aga atg ctt aca   1440
Val Asp Leu Asn Glu Thr Pro Tyr Val Gln Thr Lys Arg Met Leu Thr
465                 470                 475                 480

cgt atg aaa gcc ctc atg aaa aca ggt aaa agc tta agg aaa tgt act   1488
Arg Met Lys Ala Leu Met Lys Thr Gly Lys Ser Leu Arg Lys Cys Thr
                485                 490                 495

ttc aag ttt tat tct ctg acc aca aga ttg act gat tcg aaa ccg ttc   1536
Phe Lys Phe Tyr Ser Leu Thr Thr Arg Leu Thr Asp Ser Lys Pro Phe
            500                 505                 510

aac aac gca gtt gag aca ggt cgg aga tac ttc cca tct tgt tat gag   1584
Asn Asn Ala Val Glu Thr Gly Arg Arg Tyr Phe Pro Ser Cys Tyr Glu
        515                 520                 525

gtt ctg gat aag tac atg gat cag tat atg gac gaa gaa atc cct gat   1632
Val Leu Asp Lys Tyr Met Asp Gln Tyr Met Asp Glu Glu Ile Pro Asp
    530                 535                 540

atg tcg tat ccc gag aaa ggc act gtg aaa gag aga aga cag aag agg   1680
Met Ser Tyr Pro Glu Lys Gly Thr Val Lys Glu Arg Arg Gln Lys Arg
545                 550                 555                 560

atg aga tat aac gag ctg aag aac gac gtt aaa aaa gca tat agc aaa   1728
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Met Arg Tyr Asn Glu Leu Lys Asn Asp Val Lys Lys Ala Tyr Ser Lys
                565                 570                 575

gac aaa gtc gcg cgg tct tgt ctt tct tct tca tca cca gct tct tct   1776
Asp Lys Val Ala Arg Ser Cys Leu Ser Ser Ser Ser Pro Ala Ser Ser
            580                 585                 590

ctt aga gaa gcc tta gag aat cca aca t                            1804
Leu Arg Glu Ala Leu Glu Asn Pro Thr
        595                 600
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<212> PRT
<213> Arabidopsis thaliana

<400> 18

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Ser His Leu Ser Asn Pro Ser Pro Val Val Thr Thr Tyr His Ser Ala
             20                  25                  30

Ala Asn Leu Glu Glu Leu Ser Ser Asn Leu Glu Gln Leu Leu Thr Asn
         35                  40                  45

Pro Asp Cys Asp Tyr Thr Asp Ala Glu Ile Ile Ile Glu Glu Glu Ala
     50                  55                  60

Asn Pro Val Ser Val His Arg Cys Val Leu Ala Ala Arg Ser Lys Phe
 65                  70                  75                  80

Phe Leu Asp Leu Phe Lys Lys Asp Lys Asp Ser Ser Glu Lys Lys Pro
                 85                  90                  95

Lys Tyr Gln Met Lys Asp Leu Leu Pro Tyr Gly Asn Val Gly Arg Glu
            100                 105                 110

Ala Phe Leu His Phe Leu Ser Tyr Ile Tyr Thr Gly Arg Leu Lys Pro
        115                 120                 125

Phe Pro Ile Glu Val Ser Thr Cys Val Asp Ser Val Cys Ala His Asp
    130                 135                 140

Ser Cys Lys Pro Ala Ile Asp Phe Ala Val Glu Leu Met Tyr Ala Ser
145                 150                 155                 160

Phe Val Phe Gln Ile Pro Asp Leu Val Ser Ser Phe Gln Arg Lys Leu
                165                 170                 175

Arg Asn Tyr Val Glu Lys Ser Leu Val Glu Asn Val Leu Pro Ile Leu
            180                 185                 190

Leu Val Ala Phe His Cys Asp Leu Thr Gln Leu Leu Asp Gln Cys Ile
        195                 200                 205

Glu Arg Val Ala Arg Ser Asp Leu Asp Arg Phe Cys Ile Glu Lys Glu
    210                 215                 220

Leu Pro Leu Glu Val Leu Glu Lys Ile Lys Gln Leu Arg Val Lys Ser
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225 230 235 240

Val Asn Ile Pro Glu Val Glu Asp Lys Ser Ile Glu Arg Thr Gly Lys
245 250 255

Val Leu Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
260 265 270

Leu Thr Glu Ser Asp Ile Thr Leu Asp Gln Ala Asn Gly Leu His Tyr
275 280 285

Ala Val Ala Tyr Ser Asp Pro Lys Val Val Thr Gln Val Leu Asp Leu
290 295 300

Asp Met Ala Asp Val Asn Phe Arg Asn Ser Arg Gly Tyr Thr Val Leu
305 310 315 320

His Ile Ala Ala Met Arg Arg Glu Pro Thr Ile Ile Ile Pro Leu Ile
325 330 335

Gln Lys Gly Ala Asn Ala Ser Asp Phe Thr Phe Asp Gly Arg Ser Ala
340 345 350

Val Asn Ile Cys Arg Arg Leu Thr Arg Pro Lys Asp Tyr His Thr Lys
355 360 365

Thr Ser Arg Lys Glu Pro Ser Lys Tyr Arg Leu Cys Ile Asp Ile Leu
370 375 380

Glu Arg Glu Ile Arg Arg Asn Pro Leu Val Ser Gly Asp Thr Pro Thr
385 390 395 400

Cys Ser His Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu
405 410 415

Glu Lys Arg Trp Asp Leu Arg Gln Leu Phe Phe Pro Ala Glu Ala Asn
420 425 430

Val Ala Met Asp Val Ala Asn Val Glu Gly Thr Ser Glu Cys Thr Gly
435 440 445

Leu Leu Thr Pro Pro Pro Ser Asn Asp Thr Thr Glu Asn Leu Gly Lys
450 455 460

Val Asp Leu Asn Glu Thr Pro Tyr Val Gln Thr Lys Arg Met Leu Thr
465 470 475 480

Arg Met Lys Ala Leu Met Lys Thr Gly Lys Ser Leu Arg Lys Cys Thr
485 490 495

Phe Lys Phe Tyr Ser Leu Thr Thr Arg Leu Thr Asp Ser Lys Pro Phe
500 505 510

Asn Asn Ala Val Glu Thr Gly Arg Arg Tyr Phe Pro Ser Cys Tyr Glu
515 520 525

Val Leu Asp Lys Tyr Met Asp Gln Tyr Met Asp Glu Glu Ile Pro Asp
530 535 540

Met Ser Tyr Pro Glu Lys Gly Thr Val Lys Glu Arg Arg Gln Lys Arg
545 550 555 560

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Met Arg Tyr Asn Glu Leu Lys Asn Asp Val Lys Lys Ala Tyr Ser Lys
                565                 570                 575

Asp Lys Val Ala Arg Ser Cys Leu Ser Ser Ser Ser Pro Ala Ser Ser
            580                 585                 590

Leu Arg Glu Ala Leu Glu Asn Pro Thr
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<212> DNA
<213> Arabidopsis thaliana

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<223> AtNMLc4-2 genomic sequence

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Met Ala Thr Thr Thr Thr Thr Thr Ala Arg Phe Ser Asp Ser Tyr
  1               5                  10                  15

gag ttc agc aac aca agc ggc aat agc ttc ttc gcc gcc gag tca tct   96
Glu Phe Ser Asn Thr Ser Gly Asn Ser Phe Phe Ala Ala Glu Ser Ser
             20                  25                  30

ctt gat tat ccg acg gaa ttt ctc acg cca ccg gag gta tca gct ctt   144
Leu Asp Tyr Pro Thr Glu Phe Leu Thr Pro Pro Glu Val Ser Ala Leu
         35                  40                  45

aaa ctt ctg tct aac tgc ctc gag tct gtt ttc gac tcg ccg gag acg   192
Lys Leu Leu Ser Asn Cys Leu Glu Ser Val Phe Asp Ser Pro Glu Thr
     50                  55                  60

ttc tac agc gat gct aag cta gtt ctc gcc ggc ggc cgg gaa gtt tct   240
Phe Tyr Ser Asp Ala Lys Leu Val Leu Ala Gly Gly Arg Glu Val Ser
 65                  70                  75                  80

ttt cac cgt tgt att ctt tcc gcg aga att cct gtc ttc aaa agc gct   288
Phe His Arg Cys Ile Leu Ser Ala Arg Ile Pro Val Phe Lys Ser Ala
                 85                  90                  95

tta gcc acc gtg aag gaa caa aaa tcc tcc acc acc gtg aag ctc cag   336
Leu Ala Thr Val Lys Glu Gln Lys Ser Ser Thr Thr Val Lys Leu Gln
            100                 105                 110

ctg aaa gag atc gcc aga gat tac gaa gtc ggc ttt gac tcg gtt gtg   384
Leu Lys Glu Ile Ala Arg Asp Tyr Glu Val Gly Phe Asp Ser Val Val
        115                 120                 125

gcg gtt ttg gcg tat gtt tac agc ggc aga gtg agg tcc ccg ccg aag   432
Ala Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg Ser Pro Pro Lys
    130                 135                 140

gga gct tct gct tgc gta gac gac gat tgt tgc cac gtg gct tgc cgg   480
Gly Ala Ser Ala Cys Val Asp Asp Asp Cys Cys His Val Ala Cys Arg
```

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145                 150                 155                 160

tca aag gtg gat ttc atg gtg gag gtt ctt tat ctg tct ttc gtt ttc   528
Ser Lys Val Asp Phe Met Val Glu Val Leu Tyr Leu Ser Phe Val Phe
                165                 170                 175

cag att caa gaa tta gtt act ctg tat gag agg cag ttc ttg gaa att   576
Gln Ile Gln Glu Leu Val Thr Leu Tyr Glu Arg Gln Phe Leu Glu Ile
            180                 185                 190

gta gac aaa gtt gta gtc gaa gac atc ttg gtt ata ttc aag ctt gat   624
Val Asp Lys Val Val Val Glu Asp Ile Leu Val Ile Phe Lys Leu Asp
        195                 200                 205

act cta tgt ggt aca aca tac aag aag ctt ttg gat aga tgc ata gaa   672
Thr Leu Cys Gly Thr Thr Tyr Lys Lys Leu Leu Asp Arg Cys Ile Glu
    210                 215                 220

att atc gtg aag tct gat ata gaa cta gtt agt ctt gag aag tct tta   720
Ile Ile Val Lys Ser Asp Ile Glu Leu Val Ser Leu Glu Lys Ser Leu
225                 230                 235                 240

cct caa cac att ttc aag caa atc ata gac atc cgc gaa gcg ctc tgt   768
Pro Gln His Ile Phe Lys Gln Ile Ile Asp Ile Arg Glu Ala Leu Cys
                245                 250                 255

cta gag cca cct aaa cta gaa agg cat gtc aag aac ata tac aag gcg   816
Leu Glu Pro Pro Lys Leu Glu Arg His Val Lys Asn Ile Tyr Lys Ala
            260                 265                 270

cta gac tca gat gat gtt gag ctt gtc aag atg ctt ttg cta gaa gga   864
Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu Leu Leu Glu Gly
        275                 280                 285

cac acc aat ctc gat gag gcg tat gct ctt cat ttt gct atc gct cac   912
His Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe Ala Ile Ala His
    290                 295                 300

tgc gct gtg aag acc gcg tat gat ctc ctc gag ctt gag ctt gcg gat   960
Cys Ala Val Lys Thr Ala Tyr Asp Leu Leu Glu Leu Glu Leu Ala Asp
305                 310                 315                 320

gtt aac ctt aga aat ccg agg gga tac act gtg ctt cat gtt gct gcg   1008
Val Asn Leu Arg Asn Pro Arg Gly Tyr Thr Val Leu His Val Ala Ala
                325                 330                 335

atg cgg aag gag ccg aag ttg ata ata tct ttg tta atg aaa ggg gca   1056
Met Arg Lys Glu Pro Lys Leu Ile Ile Ser Leu Leu Met Lys Gly Ala
            340                 345                 350

aat att tta gac aca aca ttg gat ggt aga acc gct tta gtg att gta   1104
Asn Ile Leu Asp Thr Thr Leu Asp Gly Arg Thr Ala Leu Val Ile Val
        355                 360                 365

aaa cga ctc act aaa gcg gat gac tac aaa act agt acg gag gac ggt   1152
Lys Arg Leu Thr Lys Ala Asp Asp Tyr Lys Thr Ser Thr Glu Asp Gly
    370                 375                 380

acg cct tct ctg aaa ggc gga tta tgc ata gag gta ctt gag cat gaa   1200
Thr Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val Leu Glu His Glu
385                 390                 395                 400
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caa aaa cta gaa tat ttg tcg cct ata gag gct tca ctt tct ctt cca   1248
Gln Lys Leu Glu Tyr Leu Ser Pro Ile Glu Ala Ser Leu Ser Leu Pro
                405                 410                 415

gta act cca gag gag ttg agg atg agg ttg ctc tat tat gaa aac cga   1296
Val Thr Pro Glu Glu Leu Arg Met Arg Leu Leu Tyr Tyr Glu Asn Arg
            420                 425                 430

gtt gca ctt gct cga ctt ctc ttt cca gtg gaa act gaa act gta cag   1344
Val Ala Leu Ala Arg Leu Leu Phe Pro Val Glu Thr Glu Thr Val Gln
        435                 440                 445

ggt att gcc aaa ttg gag gaa aca tgc gag ttt aca gct tct agt ctc   1392
Gly Ile Ala Lys Leu Glu Glu Thr Cys Glu Phe Thr Ala Ser Ser Leu
    450                 455                 460

gag cct gat cat cac att ggt gaa aag cgg aca tca cta gac cta aat   1440
Glu Pro Asp His His Ile Gly Glu Lys Arg Thr Ser Leu Asp Leu Asn
465                 470                 475                 480

atg gcg ccg ttc caa atc cat gag aag cat ttg agt aga cta aga gca   1488
Met Ala Pro Phe Gln Ile His Glu Lys His Leu Ser Arg Leu Arg Ala
                485                 490                 495

ctt tgt aaa acc gtg gaa ctg ggg aaa cgc tac ttc aaa cga tgt tcg   1536
Leu Cys Lys Thr Val Glu Leu Gly Lys Arg Tyr Phe Lys Arg Cys Ser
            500                 505                 510

ctt gat cac ttt atg gat act gag gac ttg aat cat ctt gct agc gta   1584
Leu Asp His Phe Met Asp Thr Glu Asp Leu Asn His Leu Ala Ser Val
        515                 520                 525

gaa gaa gat act cct gag aaa cgg cta caa aag aag caa agg tac atg   1632
Glu Glu Asp Thr Pro Glu Lys Arg Leu Gln Lys Lys Gln Arg Tyr Met
    530                 535                 540

gaa cta caa gag act ctg atg aag acc ttt agt gag gac aag gag gaa   1680
Glu Leu Gln Glu Thr Leu Met Lys Thr Phe Ser Glu Asp Lys Glu Glu
545                 550                 555                 560

tgt gga aag tct tcc aca ccg aaa cca acc tct gcg gtg agg tct aat   1728
Cys Gly Lys Ser Ser Thr Pro Lys Pro Thr Ser Ala Val Arg Ser Asn
                565                 570                 575

aga aaa ctc tct cac cgg cgc cta aaa gtg gac aaa cgg gat ttt ttg   1776
Arg Lys Leu Ser His Arg Arg Leu Lys Val Asp Lys Arg Asp Phe Leu
            580                 585                 590

aaa cga cct tac ggg aac ggg gat taa                              1803
Lys Arg Pro Tyr Gly Asn Gly Asp
        595                 600
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<210> 20
<211> 600
<212> PRT
<213> Arabidopsis thaliana

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Met Ala Thr Thr Thr Thr Thr Thr Thr Ala Arg Phe Ser Asp Ser Tyr

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            20                  25                  30
Leu Asp Tyr Pro Thr Glu Phe Leu Thr Pro Pro Glu Val Ser Ala Leu
        35                  40                  45
Lys Leu Leu Ser Asn Cys Leu Glu Ser Val Phe Asp Ser Pro Glu Thr
    50                  55                  60
Phe Tyr Ser Asp Ala Lys Leu Val Leu Ala Gly Gly Arg Glu Val Ser
65                  70                  75                  80
Phe His Arg Cys Ile Leu Ser Ala Arg Ile Pro Val Phe Lys Ser Ala
                85                  90                  95
Leu Ala Thr Val Lys Glu Gln Lys Ser Ser Thr Thr Val Lys Leu Gln
            100                 105                 110
Leu Lys Glu Ile Ala Arg Asp Tyr Glu Val Gly Phe Asp Ser Val Val
        115                 120                 125
Ala Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg Ser Pro Pro Lys
    130                 135                 140
Gly Ala Ser Ala Cys Val Asp Asp Asp Cys Cys His Val Ala Cys Arg
145                 150                 155                 160
Ser Lys Val Asp Phe Met Val Glu Val Leu Tyr Leu Ser Phe Val Phe
                165                 170                 175
Gln Ile Gln Glu Leu Val Thr Leu Tyr Glu Arg Gln Phe Leu Glu Ile
            180                 185                 190
Val Asp Lys Val Val Val Glu Asp Ile Leu Val Ile Phe Lys Leu Asp
        195                 200                 205
Thr Leu Cys Gly Thr Thr Tyr Lys Lys Leu Leu Asp Arg Cys Ile Glu
    210                 215                 220
Ile Ile Val Lys Ser Asp Ile Glu Leu Val Ser Leu Glu Lys Ser Leu
225                 230                 235                 240
Pro Gln His Ile Phe Lys Gln Ile Ile Asp Ile Arg Glu Ala Leu Cys
                245                 250                 255
Leu Glu Pro Pro Lys Leu Glu Arg His Val Lys Asn Ile Tyr Lys Ala
            260                 265                 270
Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu Leu Leu Glu Gly
        275                 280                 285
His Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe Ala Ile Ala His
    290                 295                 300
Cys Ala Val Lys Thr Ala Tyr Asp Leu Leu Glu Leu Glu Leu Ala Asp
305                 310                 315                 320
Val Asn Leu Arg Asn Pro Arg Gly Tyr Thr Val Leu His Val Ala Ala
                325                 330                 335
Met Arg Lys Glu Pro Lys Leu Ile Ile Ser Leu Leu Met Lys Gly Ala
            340                 345                 350
Asn Ile Leu Asp Thr Thr Leu Asp Gly Arg Thr Ala Leu Val Ile Val
        355                 360                 365
Lys Arg Leu Thr Lys Ala Asp Asp Tyr Lys Thr Ser Thr Glu Asp Gly
    370                 375                 380
Thr Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val Leu Glu His Glu
385                 390                 395                 400
Gln Lys Leu Glu Tyr Leu Ser Pro Ile Glu Ala Ser Leu Ser Leu Pro
                405                 410                 415
Val Thr Pro Glu Glu Leu Arg Met Arg Leu Leu Tyr Tyr Glu Asn Arg
            420                 425                 430
Val Ala Leu Ala Arg Leu Leu Phe Pro Val Glu Thr Glu Thr Val Gln
        435                 440                 445
Gly Ile Ala Lys Leu Glu Glu Thr Cys Glu Phe Thr Ala Ser Ser Leu
    450                 455                 460
Glu Pro Asp His His Ile Gly Glu Lys Arg Thr Ser Leu Asp Leu Asn
465                 470                 475                 480
Met Ala Pro Phe Gln Ile His Glu Lys His Leu Ser Arg Leu Arg Ala
                485                 490                 495
```

```
Leu Cys Lys Thr Val Glu Leu Gly Lys Arg Tyr Phe Lys Arg Cys Ser
            500                 505                 510
Leu Asp His Phe Met Asp Thr Glu Asp Leu Asn His Leu Ala Ser Val
        515                 520                 525
Glu Glu Asp Thr Pro Glu Lys Arg Leu Gln Lys Lys Gln Arg Tyr Met
    530                 535                 540
Glu Leu Gln Glu Thr Leu Met Lys Thr Phe Ser Glu Asp Lys Glu Glu
545                 550                 555                 560
Cys Gly Lys Ser Ser Thr Pro Lys Pro Thr Ser Ala Val Arg Ser Asn
                565                 570                 575
Arg Lys Leu Ser His Arg Arg Leu Lys Val Asp Lys Arg Asp Phe Leu
            580                 585                 590
Lys Arg Pro Tyr Gly Asn Gly Asp
        595                 600
```

<210> 21
<211> 28
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 1A

<400> 21
gakattattg tcaagtctaa tgtwgata 28

<210> 22
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 1B

<400> 22
aytkgaytck gatgatrttg artta 25

<210> 23
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 1C

<400> 23
taaytcaaya tcatcmgart cmartgc 27

<210> 24
<211> 28
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 1D

<220>
<221> misc_feature
<222> (1)..(28)
<223> n = a, t, c or g

<400> 24
gttkagcmag nscaactcta ttttcaag 28

<210> 25
<211> 32
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 2A

<400> 25
tgcatwgara twrttgtsaa gtctratgtw ga 32

<210> 26
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 2B

<400> 26
ggcaytggay tcwgatgatg ttgaryt 27

<210> 27
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 2C

<400> 27
arytcaacat catcwgartc cartgcc 27

<210> 28
<211> 31
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 2D

<400> 28
agttkagcma gdccaactck attttcaarr t 31

<210> 29
<211> 659
<212> DNA
<213> Nicotiana tabacum

<220>
<221> CDS
<222> (1)..(657)
<223> Tobacco A

<400> 29

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tgc atg gag att att gtc aag tct aat gtt gat atc ata acc ctt gat   48
Cys Met Glu Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp
  1               5                  10                  15

aag gcc ttg cct cat gac att gta aaa caa att acc gat tca cga gca   96
Lys Ala Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala
             20                  25                  30

gaa ctt ggt cta caa ggg cct gaa agc aat ggt ttt cct gat aaa cat   144
Glu Leu Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His
         35                  40                  45

gtt aag agg ata cat agg gca tta gat tct gat gat gtt gaa tta ctg   192
Val Lys Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu
     50                  55                  60

cag atg ttg cta aga gag ggg cat act act cta gat gat gca tat gct   240
Gln Met Leu Leu Arg Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala
 65                  70                  75                  80

ctc cac tat gct gta gca tat tgc gat gca aag act aca gca gaa ctt   288
Leu His Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu
                 85                  90                  95

cta gat ctt gca ctt gct gat gtt aat cat caa aat tca aga gga tac   336
Leu Asp Leu Ala Leu Ala Asp Val Asn His Gln Asn Ser Arg Gly Tyr
            100                 105                 110

aca gtg ctg cat gtt gca gcc atg agg aaa gag cct aaa att ata gtg   384
Thr Val Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val
        115                 120                 125

tcc ctt tta acc aaa gga gct aga cct tct gat ctg aca tcc gat ggc   432
Ser Leu Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly
    130                 135                 140

aga aaa gca ctt caa att gcc aag agg ctc act agg ctt gtg gat ttc   480
Arg Lys Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe
145                 150                 155                 160

agt aag tct cca gag gaa gga aaa tct gct tcg aag gat cgg tta tgc   528
Ser Lys Ser Pro Glu Glu Gly Lys Ser Ala Ser Lys Asp Arg Leu Cys
                165                 170                 175

att gag att ctg gag caa gca gaa aga aga gat cca ctg cta gga gaa   576
Ile Glu Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu
```

```
            180                 185                 190

gct tct gta tct ctt gct atg gcg ggc gat gat ttg cgt atg aag ctg   624
Ala Ser Val Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu
        195                 200                 205

tta tac ctt gaa aat aga gtt ggc ctt gct caa ct                    659
Leu Tyr Leu Glu Asn Arg Val Gly Leu Ala Gln
    210                 215
```

<210> 30
<211> 219
<212> PRT
<213> Nicotiana tabacum

<400> 30

```
Cys Met Glu Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp
  1               5                  10                  15

Lys Ala Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala
             20                  25                  30

Glu Leu Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His
         35                  40                  45

Val Lys Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu
     50                  55                  60

Gln Met Leu Leu Arg Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala
 65                  70                  75                  80

Leu His Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu
                 85                  90                  95

Leu Asp Leu Ala Leu Ala Asp Val Asn His Gln Asn Ser Arg Gly Tyr
            100                 105                 110

Thr Val Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val
        115                 120                 125

Ser Leu Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly
    130                 135                 140

Arg Lys Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe
145                 150                 155                 160

Ser Lys Ser Pro Glu Glu Gly Lys Ser Ala Ser Lys Asp Arg Leu Cys
                165                 170                 175

Ile Glu Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu
            180                 185                 190

Ala Ser Val Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu
        195                 200                 205

Leu Tyr Leu Glu Asn Arg Val Gly Leu Ala Gln
    210                 215
```

<210> 31
<211> 498
<212> DNA
<213> Nicotiana tabacum

<220>
<221> CDS
<222> (2)..(496)
<223> Tobacco B

<400> 31
g gca ctg gat tct gat gat gtt gag ctg gtc aag ctt cta ctc aac gag 49
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu Leu Asn Glu
1 5 10 15

tct gag ata agc tta gat gaa gcc tac gct ctt cat tat gct gtt gca 97
Ser Glu Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

tat tgt gat ccc aag gtt gtg act gag gtt ctt gga ctg ggt gtt gct 145
Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
35 40 45

gat gtc aat cta cgt aat act cgc ggt tac act gtg ctt cac att gct 193
Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
50 55 60

gcc atg cgt aag gag cca gca ata att gta tcg ctt ttg act aag gga 241
Ala Met Arg Lys Glu Pro Ala Ile Ile Val Ser Leu Leu Thr Lys Gly
65 70 75 80

gct cat gtg tca gag att aca ttg gat ggg caa agt gct gtt agt atc 289
Ala His Val Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
85 90 95

tgt agg agg cta act agg cct aag gag tac cat gca aaa aca gaa caa 337
Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
100 105 110

ggc cag gaa gca aac aaa gat cgg gta tgt att gat gtt ttg gag aga 385
Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
115 120 125

gag atg cgt cgc aac cca atg gct gga gat gca ttg ctt tct tcc caa 433
Glu Met Arg Arg Asn Pro Met Ala Gly Asp Ala Leu Leu Ser Ser Gln
130 135 140

atg ttg gcc gat gat ctg cac atg aaa ctg cac tat ttt gaa aat cga 481
Met Leu Ala Asp Asp Leu His Met Lys Leu His Tyr Phe Glu Asn Arg
145 150 155 160

gtt gga ctt gct caa ct 498
Val Gly Leu Ala Gln
165

<210> 32
<211> 165
<212> PRT
<213> Nicotiana tabacum

<400> 32
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu Leu Asn Glu
1 5 10 15

Ser Glu Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
35 40 45

Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
50 55 60

Ala Met Arg Lys Glu Pro Ala Ile Ile Val Ser Leu Leu Thr Lys Gly
65 70 75 80

Ala His Val Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
85 90 95

Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
100 105 110

Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
115 120 125

Glu Met Arg Arg Asn Pro Met Ala Gly Asp Ala Leu Leu Ser Ser Gln
130 135 140

Met Leu Ala Asp Asp Leu His Met Lys Leu His Tyr Phe Glu Asn Arg
145 150 155 160

Val Gly Leu Ala Gln
165

<210> 33
<211> 498
<212> DNA
<213> Nicotiana tabacum

<220>
<221> CDS
<222> (2)..(496)
<223> Tobacco C

<400> 33
g gca ctg gac tcw gat gat gtt gag ttt gtc aag ctt cta ctg agt gag 49
Ala Leu Asp Xaa Asp Asp Val Glu Phe Val Lys Leu Leu Leu Ser Glu
1 5 10 15

tct aac ata agc tta gat gaa gcc tac gct ctt cat tat gct gtg gca 97
Ser Asn Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

tat tgt gat ccc aag gtt gtg act gag gtt ctt gga ctg ggt gtt gcg 145
Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
35 40 45

gat gtc aac cta cgt aat act cgt ggt tac act gtg ctt cac att gct 193
Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala

```
     50                  55                  60

tcc atg cgt aag gag cca gca gta att gta tcg ctt ttg act aag gga   241
Ser Met Arg Lys Glu Pro Ala Val Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

gct cgt gca tca gag act aca ttg gat ggg cag agt gct gtt agt atc   289
Ala Arg Ala Ser Glu Thr Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95

tgt agg agg ctg act agg cct aag gag tac cat gca aaa aca gaa caa   337
Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
            100                 105                 110

ggc cag gaa gca aac aaa gat cgg gta tgt att gat gtt ttg gag aga   385
Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
        115                 120                 125

gag atg cgt cgc aac cca atg gct gga gat gca ttg ttt tct tcc cca   433
Glu Met Arg Arg Asn Pro Met Ala Gly Asp Ala Leu Phe Ser Ser Pro
    130                 135                 140

atg ttg gcc gat gat ctg cac atg aaa ctg cac tac ctt gaa aat aga   481
Met Leu Ala Asp Asp Leu His Met Lys Leu His Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggc ctg gct caa ct                                           498
Val Gly Leu Ala Gln
                165
```

<210> 34
<211> 165
<212> PRT
<213> Nicotiana tabacum

```
<400> 34
Ala Leu Asp Xaa Asp Asp Val Glu Phe Val Lys Leu Leu Leu Ser Glu
  1               5                  10                  15

Ser Asn Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr Ala Val Ala
             20                  25                  30

Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
         35                  40                  45

Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
     50                  55                  60

Ser Met Arg Lys Glu Pro Ala Val Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala Arg Ala Ser Glu Thr Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95

Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
            100                 105                 110

Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
        115                 120                 125
```

```
Glu Met Arg Arg Asn Pro Met Ala Gly Asp Ala Leu Phe Ser Ser Pro
    130                 135                 140

Met Leu Ala Asp Asp Leu His Met Lys Leu His Tyr Leu Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Gln
                165
```

<210> 35
<211> 399
<212> DNA
<213> Nicotiana tabacum

<220>
<221> CDS
<222> (1)..(399)
<223> Tobacco D

<400> 35

```
act gat tcg gat gat gtt gag tta ctt aag tta ctt ctt gaa gag tct   48
Thr Asp Ser Asp Asp Val Glu Leu Leu Lys Leu Leu Leu Glu Glu Ser
  1               5                  10                  15

aat gtc act tta gac gat gct tgt gct ctt cat tat gca gct gct tat   96
Asn Val Thr Leu Asp Asp Ala Cys Ala Leu His Tyr Ala Ala Ala Tyr
             20                  25                  30

tgt aac tcc aag gtt gtg aat gag gtc ctc gag ctg gat tta gct gat   144
Cys Asn Ser Lys Val Val Asn Glu Val Leu Glu Leu Asp Leu Ala Asp
         35                  40                  45

gtc aat ctt cag aac tcc cga gga tat aac gtc ctt cac gtt gct gct   192
Val Asn Leu Gln Asn Ser Arg Gly Tyr Asn Val Leu His Val Ala Ala
     50                  55                  60

aga aga aag gag cca tca ata ata atg gga cta ctt gaa aaa gga gca   240
Arg Arg Lys Glu Pro Ser Ile Ile Met Gly Leu Leu Glu Lys Gly Ala
 65                  70                  75                  80

tct ttc ttg aat act aca cgg gat gga aac aca gca cta tct atc tgt   288
Ser Phe Leu Asn Thr Thr Arg Asp Gly Asn Thr Ala Leu Ser Ile Cys
                 85                  90                  95

cgg aga ttg act cgg cca aag gat tat aat gag cca aca aag caa ggg   336
Arg Arg Leu Thr Arg Pro Lys Asp Tyr Asn Glu Pro Thr Lys Gln Gly
            100                 105                 110

aaa gaa act aat aag gac cgc ata tgc att gat att ttg gag aga gag   384
Lys Glu Thr Asn Lys Asp Arg Ile Cys Ile Asp Ile Leu Glu Arg Glu
        115                 120                 125

acg aat agg aat cct                                              399
Thr Asn Arg Asn Pro
    130
```

<210> 36
<211> 133

<212> PRT
<213> Nicotiana tabacum

<400> 36
Thr Asp Ser Asp Asp Val Glu Leu Leu Lys Leu Leu Leu Glu Glu Ser
1 5 10 15

Asn Val Thr Leu Asp Asp Ala Cys Ala Leu His Tyr Ala Ala Ala Tyr
20 25 30

Cys Asn Ser Lys Val Val Asn Glu Val Leu Glu Leu Asp Leu Ala Asp
35 40 45

Val Asn Leu Gln Asn Ser Arg Gly Tyr Asn Val Leu His Val Ala Ala
50 55 60

Arg Arg Lys Glu Pro Ser Ile Ile Met Gly Leu Leu Glu Lys Gly Ala
65 70 75 80

Ser Phe Leu Asn Thr Thr Arg Asp Gly Asn Thr Ala Leu Ser Ile Cys
85 90 95

Arg Arg Leu Thr Arg Pro Lys Asp Tyr Asn Glu Pro Thr Lys Gln Gly
100 105 110

Lys Glu Thr Asn Lys Asp Arg Ile Cys Ile Asp Ile Leu Glu Arg Glu
115 120 125

Thr Asn Arg Asn Pro
130

<210> 37
<211> 498
<212> DNA
<213> Lycopersicon esculentum

<220>
<221> CDS
<222> (2)..(496)
<223> Tomato A

<400> 37
g gca ttg gat tct gat gat gtt gag tta cta agg atg ttg ctt aaa gag 49
Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met Leu Leu Lys Glu
1 5 10 15

ggg cat act act ctt gat gat gca tat gct ctc cac tat gct gta gca 97
Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

tat tgc gat gca aag act aca gca gaa ctt tta gat ctt tca ctt gct 145
Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp Leu Ser Leu Ala
35 40 45

gat gtt aat cat caa aat cct aga gga cac acg gta ctt cat gtt gct 193
Asp Val Asn His Gln Asn Pro Arg Gly His Thr Val Leu His Val Ala
50 55 60

gcc atg agg aaa gaa cct aaa att ata gtg tcc ctt tta acc aaa gga 241

```
Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

gct aga cct tct gat ctg aca tcc gat ggc aaa aaa gca ctt caa att   289
Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys Ala Leu Gln Ile
                85                  90                  95

gct aag agg ctc act agg ctt gta gat ttt acc aag tct aca gag gaa   337
Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys Ser Thr Glu Glu
            100                 105                 110

gga aaa tct gct cca aag gat cgg tta tgc att gag att ctg gag caa   385
Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln
        115                 120                 125

gca gaa aga aga gat cca cta cta gga gaa gct tca tta tct ctt gct   433
Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser Leu Ser Leu Ala
    130                 135                 140

atg gca ggc gat gat ttg cgt atg aag ctg tta tac ctt gaa aat aga   481
Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggc ctt gct aaa ct                                           498
Val Gly Leu Ala Lys
                165
```

<210> 38
<211> 165
<212> PRT
<213> Lycopersicon esculentum

<400> 38

```
Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met Leu Leu Lys Glu
  1               5                  10                  15

Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
             20                  25                  30

Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp Leu Ser Leu Ala
         35                  40                  45

Asp Val Asn His Gln Asn Pro Arg Gly His Thr Val Leu His Val Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys Ala Leu Gln Ile
                85                  90                  95

Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys Ser Thr Glu Glu
            100                 105                 110

Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln
        115                 120                 125

Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser Leu Ser Leu Ala
    130                 135                 140
```

```
Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Lys
                165
```

<210> 39
<211> 498
<212> DNA
<213> Beta vulgaris

<220>
<221> CDS
<222> (2)..(496)
<223> Sugarbeet

<400> 39

```
g gca ttg gat tct gat gat gtt gag tta gtc aga atg ctt tta aaa gag 49
  Ala Leu Asp Ser Asp Asp Val Glu Leu Val Arg Met Leu Leu Lys Glu
    1               5                  10                  15

cgc cat aca act cta gat gat gca tat gcc ctt cac tat gct gtg gca   97
Arg His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
            20                  25                  30

cat tgt gat gcc aag acc acc acg gag ctt ctt gag ctt ggg ctt gca   145
His Cys Asp Ala Lys Thr Thr Thr Glu Leu Leu Glu Leu Gly Leu Ala
        35                  40                  45

gat gtt aat ctt aga aat cta agg ggt cac act gtg cta cat gtg gca   193
Asp Val Asn Leu Arg Asn Leu Arg Gly His Thr Val Leu His Val Ala
    50                  55                  60

gcc atg aga aaa gag cct aag ata att gta tcc ttg tta acc aag gga   241
Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu Leu Thr Lys Gly
65                  70                  75                  80

gcc cat ccg tct gat ata aca tca gat gat aaa aaa gca ctg cag ata   289
Ala His Pro Ser Asp Ile Thr Ser Asp Asp Lys Lys Ala Leu Gln Ile
                85                  90                  95

gca aag aga cta aca aaa gct gtg gac ttc tat aaa act aca gaa caa   337
Ala Lys Arg Leu Thr Lys Ala Val Asp Phe Tyr Lys Thr Thr Glu Gln
            100                 105                 110

gga aaa gat gca cca aag gat cgg ttg tgc att gaa ata ctg gag caa   385
Gly Lys Asp Ala Pro Lys Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln
        115                 120                 125

gct gaa aga aga gaa cca ttg cta gga gaa ggt tct gtt tct ctt gca   433
Ala Glu Arg Arg Glu Pro Leu Leu Gly Glu Gly Ser Val Ser Leu Ala
    130                 135                 140

aag gca gga gat gat ctg cgt atg aag cta tta tac ctt gaa aat cga   481
Lys Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggc ctt gct caa ct                                           498
Val Gly Leu Ala Gln
```

165

<210> 40
<211> 165
<212> PRT
<213> Beta vulgaris

<400> 40
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Arg Met Leu Leu Lys Glu
1 5 10 15

Arg His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

His Cys Asp Ala Lys Thr Thr Thr Glu Leu Leu Glu Leu Gly Leu Ala
35 40 45

Asp Val Asn Leu Arg Asn Leu Arg Gly His Thr Val Leu His Val Ala
50 55 60

Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu Leu Thr Lys Gly
65 70 75 80

Ala His Pro Ser Asp Ile Thr Ser Asp Asp Lys Lys Ala Leu Gln Ile
85 90 95

Ala Lys Arg Leu Thr Lys Ala Val Asp Phe Tyr Lys Thr Thr Glu Gln
100 105 110

Gly Lys Asp Ala Pro Lys Asp Arg Leu Cys Ile Glu Ile Leu Glu Gln
115 120 125

Ala Glu Arg Arg Glu Pro Leu Leu Gly Glu Gly Ser Val Ser Leu Ala
130 135 140

Lys Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu Asn Arg
145 150 155 160

Val Gly Leu Ala Gln
165

<210> 41
<211> 498
<212> DNA
<213> Helianthus annuus

<220>
<221> CDS
<222> (2)..(496)
<223> Sunflower A

<400> 41
g gca ttg gat tct gat gat gtt gag yta gtc aca atg tta tta cga gaa 49
Ala Leu Asp Ser Asp Asp Val Glu Xaa Val Thr Met Leu Leu Arg Glu
1 5 10 15

ggt cat act tca tta gac ggt tct tgc gct ctt cat tac gct gtt gcg 97
Gly His Thr Ser Leu Asp Gly Ser Cys Ala Leu His Tyr Ala Val Ala

```
             20                  25                  30

tac gca gat gct aaa acg aca acc gaa tta ctg gat tta gca ctt gct   145
Tyr Ala Asp Ala Lys Thr Thr Thr Glu Leu Leu Asp Leu Ala Leu Ala
         35                  40                  45

gac gta aat cat aaa aac tcg agg ggt ttt acc gta ctt cat gtt gcc   193
Asp Val Asn His Lys Asn Ser Arg Gly Phe Thr Val Leu His Val Ala
     50                  55                  60

gct atg aga aaa gag ccg agt att atc gtt tcg ctt ctt acg aaa ggg   241
Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

gcc cga ccc tcg gat ctc acc cct gat ggg aga aaa gca cta cag att   289
Ala Arg Pro Ser Asp Leu Thr Pro Asp Gly Arg Lys Ala Leu Gln Ile
                 85                  90                  95

tcg aag agg ttg acc aga gcg gtt gac tat tac aag tca aac gag gat   337
Ser Lys Arg Leu Thr Arg Ala Val Asp Tyr Tyr Lys Ser Asn Glu Asp
            100                 105                 110

gat aaa gag tca acg aaa ggt cgt ttg tgt att gag ata ttg gaa caa   385
Asp Lys Glu Ser Thr Lys Gly Arg Leu Cys Ile Glu Ile Leu Glu Gln
        115                 120                 125

gcc gaa aga aga aat cca ttg tta ggt gaa gct tcg gct tct ctt gca   433
Ala Glu Arg Arg Asn Pro Leu Leu Gly Glu Ala Ser Ala Ser Leu Ala
    130                 135                 140

atg gcc gga gat gat ttg cgt gga aag ttg ttg tac ctt gaa aat cga   481
Met Ala Gly Asp Asp Leu Arg Gly Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggc ctg gct caa ct                                           498
Val Gly Leu Ala Gln
                165
```

<210> 42
<211> 165
<212> PRT
<213> Helianthus annuus

<400> 42

```
Ala Leu Asp Ser Asp Asp Val Glu Xaa Val Thr Met Leu Leu Arg Glu
  1               5                  10                  15

Gly His Thr Ser Leu Asp Gly Ser Cys Ala Leu His Tyr Ala Val Ala
             20                  25                  30

Tyr Ala Asp Ala Lys Thr Thr Thr Glu Leu Leu Asp Leu Ala Leu Ala
         35                  40                  45

Asp Val Asn His Lys Asn Ser Arg Gly Phe Thr Val Leu His Val Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala Arg Pro Ser Asp Leu Thr Pro Asp Gly Arg Lys Ala Leu Gln Ile
```

```
                85                  90                  95

Ser Lys Arg Leu Thr Arg Ala Val Asp Tyr Tyr Lys Ser Asn Glu Asp
            100                 105                 110

Asp Lys Glu Ser Thr Lys Gly Arg Leu Cys Ile Glu Ile Leu Glu Gln
        115                 120                 125

Ala Glu Arg Arg Asn Pro Leu Leu Gly Glu Ala Ser Ala Ser Leu Ala
    130                 135                 140

Met Ala Gly Asp Asp Leu Arg Gly Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Gln
                165
```

<210> 43
<211> 498
<212> DNA
<213> Helianthus annuus

<220>
<221> CDS
<222> (2)..(496)
<223> Sunflower B

<400> 43

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g gca ttg gac tct gat gat gtt gag ctt gtg aaa atg att tta gac gaa 49
  Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Ile Leu Asp Glu
    1               5                  10                  15

tcc aaa atc acg tta gat gaa gcc tgc gct ctt cat tat gcg gtc atg   97
Ser Lys Ile Thr Leu Asp Glu Ala Cys Ala Leu His Tyr Ala Val Met
             20                  25                  30

tat tgt aat caa gaa gtt gct aag gag att ctt aac tta aac cgt gcg   145
Tyr Cys Asn Gln Glu Val Ala Lys Glu Ile Leu Asn Leu Asn Arg Ala
         35                  40                  45

gat gtt aat ctt aga aac tca cga gat tac acc gtg ctt cat gtt gct   193
Asp Val Asn Leu Arg Asn Ser Arg Asp Tyr Thr Val Leu His Val Ala
     50                  55                  60

gcc atg cgt aaa gaa cca tca ctt att gtt tcg att cta agc aaa ggc   241
Ala Met Arg Lys Glu Pro Ser Leu Ile Val Ser Ile Leu Ser Lys Gly
 65                  70                  75                  80

gcg tgt gca tcg gat act act ttt gat gga caa agt gcg gtt agt att   289
Ala Cys Ala Ser Asp Thr Thr Phe Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95

tgc agg aga cga aca agg ccc aag gat tat tat gtg aaa acc gaa cac   337
Cys Arg Arg Arg Thr Arg Pro Lys Asp Tyr Tyr Val Lys Thr Glu His
            100                 105                 110

ggg caa gaa aca aat aaa gat cgt ata tgc atc gat gtt ttg gag cgg   385
Gly Gln Glu Thr Asn Lys Asp Arg Ile Cys Ile Asp Val Leu Glu Arg
        115                 120                 125
```

```
gaa ata aag agg aat ccg atg ata ggc gat gtt tcc gtg tgt tct tca   433
Glu Ile Lys Arg Asn Pro Met Ile Gly Asp Val Ser Val Cys Ser Ser
    130                 135                 140

gca gtg gct gat gat ttg cat atg aat tta ctc tac ttt gaa aat cga   481
Ala Val Ala Asp Asp Leu His Met Asn Leu Leu Tyr Phe Glu Asn Arg
145                 150                 155                 160

gtt ggc ctt gct caa ct                                           498
Val Gly Leu Ala Gln
                165
```

<210> 44
<211> 165
<212> PRT
<213> Helianthus annuus

<400> 44

```
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Ile Leu Asp Glu
  1               5                  10                  15

Ser Lys Ile Thr Leu Asp Glu Ala Cys Ala Leu His Tyr Ala Val Met
             20                  25                  30

Tyr Cys Asn Gln Glu Val Ala Lys Glu Ile Leu Asn Leu Asn Arg Ala
         35                  40                  45

Asp Val Asn Leu Arg Asn Ser Arg Asp Tyr Thr Val Leu His Val Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Leu Ile Val Ser Ile Leu Ser Lys Gly
 65                  70                  75                  80

Ala Cys Ala Ser Asp Thr Thr Phe Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95

Cys Arg Arg Arg Thr Arg Pro Lys Asp Tyr Tyr Val Lys Thr Glu His
            100                 105                 110

Gly Gln Glu Thr Asn Lys Asp Arg Ile Cys Ile Asp Val Leu Glu Arg
        115                 120                 125

Glu Ile Lys Arg Asn Pro Met Ile Gly Asp Val Ser Val Cys Ser Ser
    130                 135                 140

Ala Val Ala Asp Asp Leu His Met Asn Leu Leu Tyr Phe Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Gln
                165
```

<210> 45
<211> 653
<212> DNA
<213> Solanum tuberosum

<220>

<221> CDS
<222> (1)..(651)
<223> Potato A

<400> 45

```
gak att att gtc aag tct aat gtt gat atc ata acc ctt gat aag tcc   48
Xaa Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ser
  1               5                  10                  15

ttg cct cat gac atc gta aaa caa atc act gat tca cgt gct gaa ctt   96
Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu
            20                  25                  30

ggt cta caa ggg cct gaa agc aat ggt ttt cct gat aaa cat gtt aag   144
Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys
        35                  40                  45

agg ata cat agg gca ttg gac tct gat gat gtt gag tta cta agg atg   192
Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met
    50                  55                  60

ttg ctt aaa gaa ggg cat act act ctc gat gat gca tat gct ctc cac   240
Leu Leu Lys Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His
65                  70                  75                  80

tat gct gta gca tat tgc gat gca aag act aca gca gaa ctt tta gat   288
Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp
                85                  90                  95

ctt tca ctt gct gat gtt aat cat caa aat cct aga gga tac acg gta   336
Leu Ser Leu Ala Asp Val Asn His Gln Asn Pro Arg Gly Tyr Thr Val
            100                 105                 110

ctt cat gtt gct gcc atg agg aaa gag cct aaa att ata gtg tcc ctt   384
Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu
        115                 120                 125

tta acc aaa gga gct aga cct tct gat ctg aca tct gat ggc aaa aaa   432
Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys
    130                 135                 140

gca ctt caa att gct aag agg ctc act agg ctt gtg gat ttt act aag   480
Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys
145                 150                 155                 160

tct aca gag gaa gga aaa tct gct cca aaa gat cgg tta tgc att gag   528
Ser Thr Glu Glu Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu
                165                 170                 175

att ctg gag caa gca gaa aga aga gat cca cta cta gga gaa gct tca   576
Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser
            180                 185                 190

tta tct ctt gct atg gca ggc gat gat ttg cgt atg aag ctg tta tac   624
Leu Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr
        195                 200                 205

ctt gaa aat cga gtt ggc ctk gct caa ct                           653
Leu Glu Asn Arg Val Gly Xaa Ala Gln
    210                 215
```

<210> 46
<211> 217
<212> PRT
<213> Solanum tuberosum

<400> 46
Xaa Ile Ile Val Lys Ser Asn Val Asp Ile Ile Thr Leu Asp Lys Ser
1 5 10 15

Leu Pro His Asp Ile Val Lys Gln Ile Thr Asp Ser Arg Ala Glu Leu
20 25 30

Gly Leu Gln Gly Pro Glu Ser Asn Gly Phe Pro Asp Lys His Val Lys
35 40 45

Arg Ile His Arg Ala Leu Asp Ser Asp Asp Val Glu Leu Leu Arg Met
50 55 60

Leu Leu Lys Glu Gly His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His
65 70 75 80

Tyr Ala Val Ala Tyr Cys Asp Ala Lys Thr Thr Ala Glu Leu Leu Asp
85 90 95

Leu Ser Leu Ala Asp Val Asn His Gln Asn Pro Arg Gly Tyr Thr Val
100 105 110

Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu
115 120 125

Leu Thr Lys Gly Ala Arg Pro Ser Asp Leu Thr Ser Asp Gly Lys Lys
130 135 140

Ala Leu Gln Ile Ala Lys Arg Leu Thr Arg Leu Val Asp Phe Thr Lys
145 150 155 160

Ser Thr Glu Glu Gly Lys Ser Ala Pro Lys Asp Arg Leu Cys Ile Glu
165 170 175

Ile Leu Glu Gln Ala Glu Arg Arg Asp Pro Leu Leu Gly Glu Ala Ser
180 185 190

Leu Ser Leu Ala Met Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr
195 200 205

Leu Glu Asn Arg Val Gly Xaa Ala Gln
210 215

<210> 47
<211> 498
<212> DNA
<213> Solanum tuberosum

<220>
<221> CDS
<222> (2)..(496)
<223> Potato B

<400> 47
g gca ttg gat tca gat gat gtt gag ttt gtc aag ctt cta ctt aat gag 49
Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Asn Glu
1 5 10 15

tct gac ata agt tta gat gga gcc tac gct ctt cat tac gct gtt gca 97
Ser Asp Ile Ser Leu Asp Gly Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

tat tgt gac ccc aag gtt gtt act gag gtt ctt gga ctg ggt gtt gct 145
Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
35 40 45

aat gtc aac ctt cgg aat aca cgt ggt tac act gtg ctt cac att gct 193
Asn Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
50 55 60

gcc atg cgt aag gaa ccc tca atc att gta tca ctt ttg act aag gga 241
Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
65 70 75 80

gct cat gca tca gaa att aca ttg gat ggg cag agt gct gtt ggc atc 289
Ala His Ala Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Gly Ile
85 90 95

tgt agg agg ctg agt agg cct aag gag tac cat gca aaa aca gaa caa 337
Cys Arg Arg Leu Ser Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
100 105 110

ggc cag gaa gca aac aaa gat cgg gta tgt att gat gtt ttg gag aga 385
Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
115 120 125

gag atg cgt cac aac cca atg acc gga gat gca tta ttt tct tcc ccc 433
Glu Met Arg His Asn Pro Met Thr Gly Asp Ala Leu Phe Ser Ser Pro
130 135 140

atg ttg gcc gat gat ctg ccc atg aaa ctg ctc tac ctt gaa aat cga 481
Met Leu Ala Asp Asp Leu Pro Met Lys Leu Leu Tyr Leu Glu Asn Arg
145 150 155 160

gtt ggc ctt gct aaa ct 498
Val Gly Leu Ala Lys
165

<210> 48
<211> 165
<212> PRT
<213> Solanum tuberosum

<400> 48
Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Asn Glu
1 5 10 15

Ser Asp Ile Ser Leu Asp Gly Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
35 40 45

```
Asn Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala His Ala Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Gly Ile
                 85                  90                  95

Cys Arg Arg Leu Ser Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
            100                 105                 110

Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
        115                 120                 125

Glu Met Arg His Asn Pro Met Thr Gly Asp Ala Leu Phe Ser Ser Pro
    130                 135                 140

Met Leu Ala Asp Asp Leu Pro Met Lys Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Lys
                165
```

<210> 49
<211> 477
<212> DNA
<213> Solanum tuberosum

<220>
<221> CDS
<222> (2)..(475)
<223> Potato C

<400> 49

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g gca ctg gac tct gat gat gtt gag ttt gtc aag ctt cta ctt aat gag 49
  Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Asn Glu
    1               5                  10                  15

tct gac ata agt tta gat gga gcc tac gct ctt cat tac gct gtt gca   97
Ser Asp Ile Ser Leu Asp Gly Ala Tyr Ala Leu His Tyr Ala Val Ala
             20                  25                  30

tat tgt gac ccc aag gtt gtt act gag gtt ctt gga ctg ggt gtt gct   145
Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
         35                  40                  45

aat gtc aac ctt cgg aat aca cgt ggt tac act gtg ctt cac att gct   193
Asn Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
     50                  55                  60

gcc atg cgt aag gaa ccc tca atc att gta tca ctt ttg act aag gga   241
Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

gct cat gca tca gaa att aca ttg gat ggg cag agt gct gtt agc atc   289
Ala His Ala Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95
```

```
tgt agg agg ctg act agg cct aag gag tac cat gca aaa aca gaa caa   337
Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
            100                 105                 110

ggc cag gaa gca aac aaa gat cgg gta tgt att gat gtt ttg gag aga   385
Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
        115                 120                 125

gag atg cgt cgc aac cca atg acc gga gat gca tta ttt tct tcc ccc   433
Glu Met Arg Arg Asn Pro Met Thr Gly Asp Ala Leu Phe Ser Ser Pro
    130                 135                 140

atg aaa cag ctc tac ctt gaa aat aga gtt ggc ctt gct aaa ct        477
Met Lys Gln Leu Tyr Leu Glu Asn Arg Val Gly Leu Ala Lys
145                 150                 155
```

<210> 50
<211> 158
<212> PRT
<213> Solanum tuberosum

<400> 50

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Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Asn Glu
  1               5                  10                  15

Ser Asp Ile Ser Leu Asp Gly Ala Tyr Ala Leu His Tyr Ala Val Ala
             20                  25                  30

Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu Gly Val Ala
         35                  40                  45

Asn Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu His Ile Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Ile Ile Val Ser Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala His Ala Ser Glu Ile Thr Leu Asp Gly Gln Ser Ala Val Ser Ile
                 85                  90                  95

Cys Arg Arg Leu Thr Arg Pro Lys Glu Tyr His Ala Lys Thr Glu Gln
            100                 105                 110

Gly Gln Glu Ala Asn Lys Asp Arg Val Cys Ile Asp Val Leu Glu Arg
        115                 120                 125

Glu Met Arg Arg Asn Pro Met Thr Gly Asp Ala Leu Phe Ser Ser Pro
    130                 135                 140

Met Lys Gln Leu Tyr Leu Glu Asn Arg Val Gly Leu Ala Lys
145                 150                 155
```

<210> 51
<211> 501
<212> DNA
<213> Brassica napus

<220>

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<221> CDS
<222> (2)..(499)
<223> Canola A

<400> 51
g gca ttg gat tct gat gat gtt gag ttt gtg aag ttg ctt ttg act gag 49
  Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Thr Glu
    1               5                  10                  15

tca gat atc act cta gat gaa gcc aat ggt ctt cat tac tca gtg gtg   97
Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
            20                  25                  30

tat agt gat ccc aaa gtt gtt gcc gag att ctt act ctt gat atg ggt   145
Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Thr Leu Asp Met Gly
        35                  40                  45

gat gtc aac cac aga aac tca cgt ggc tac acg gtt ctt cat ctc gca   193
Asp Val Asn His Arg Asn Ser Arg Gly Tyr Thr Val Leu His Leu Ala
    50                  55                  60

gcc atg cgc aaa gag ccg tcc atc atc ata tct ctt ctc aag aga ggt   241
Ala Met Arg Lys Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Arg Gly
65                  70                  75                  80

gcc aat gcg tct ggc ttc acg tgt gat gga cgc agt gcg gtt aat ata   289
Ala Asn Ala Ser Gly Phe Thr Cys Asp Gly Arg Ser Ala Val Asn Ile
                85                  90                  95

tgt aga aga ttg aca act cca aag gat tat cat acg aaa aca gct gcg   337
Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ala Ala
            100                 105                 110

aaa ggg agg gaa gct agt aaa gca cgg tta tgt ata gat ctc ttg gaa   385
Lys Gly Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu
        115                 120                 125

aga gaa gta agg agg aac cct atg gtt gtt gat tca cca atg tgt tcc   433
Arg Glu Val Arg Arg Asn Pro Met Val Val Asp Ser Pro Met Cys Ser
    130                 135                 140

ctt tct atg cct gaa gat ctc caa atg aga ctg tta tac ctt gaa aat   481
Leu Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn
145                 150                 155                 160

cga gtt ggc ctt gct caa ct                                       501
Arg Val Gly Leu Ala Gln
                165


<210> 52
<211> 166
<212> PRT
<213> Brassica napus

<400> 52
Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Thr Glu
  1               5                  10                  15

Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
            20                  25                  30
```

```
Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Thr Leu Asp Met Gly
        35                  40                  45

Asp Val Asn His Arg Asn Ser Arg Gly Tyr Thr Val Leu His Leu Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Arg Gly
 65                  70                  75                  80

Ala Asn Ala Ser Gly Phe Thr Cys Asp Gly Arg Ser Ala Val Asn Ile
                85                  90                  95

Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ala Ala
            100                 105                 110

Lys Gly Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu
        115                 120                 125

Arg Glu Val Arg Arg Asn Pro Met Val Val Asp Ser Pro Met Cys Ser
    130                 135                 140

Leu Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn
145                 150                 155                 160

Arg Val Gly Leu Ala Gln
                165
```

<210> 53
<211> 501
<212> DNA
<213> Brassica napus

<220>
<221> CDS
<222> (2)..(499)
<223> Canola B

<400> 53

```
g gca ttg gat tct gat gat gtt gag ttt gtg aag ctt ctt ttg acc gag 49
  Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Thr Glu
    1               5                  10                  15

tca gat atc act cta gat gaa gcc aat ggt ctt cat tac tca gtg gtg   97
Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
            20                  25                  30

tat agt gat ccc aaa gtt gtt gcc gag att ctt act ctt gat atg ggt   145
Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Thr Leu Asp Met Gly
        35                  40                  45

gat gtt aac cac aga aac tca cgt ggc tac acg gtt ctg cat ctc gca   193
Asp Val Asn His Arg Asn Ser Arg Gly Tyr Thr Val Leu His Leu Ala
     50                  55                  60

gcc atg cgc aaa gag ccg tcc atc atc ata tct ctt ctc aag aaa ggt   241
Ala Met Arg Lys Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Lys Gly
 65                  70                  75                  80
```

```
gcc aat gcg tct ggc ttc acc tgt gat gga cgc agt gcg gtt aat ata   289
Ala Asn Ala Ser Gly Phe Thr Cys Asp Gly Arg Ser Ala Val Asn Ile
                 85                  90                  95

tgt aga aga ttg aca act cca aag gat tat cat act aaa aca gct gcg   337
Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ala Ala
            100                 105                 110

aaa ggg agg gaa gct agt aaa gca cgg tta tgt ata gat ctc ttg gaa   385
Lys Gly Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu
        115                 120                 125

aga gaa gta agg agg aac cct atg gtt gtt gag tca cca atg tgt tct   433
Arg Glu Val Arg Arg Asn Pro Met Val Val Glu Ser Pro Met Cys Ser
    130                 135                 140

ctt tct atg cct gaa gat ctc caa atg aga ctg tta tac ctt gaa aat   481
Leu Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn
145                 150                 155                 160

cga gtt ggc ctg gct caa ct                                       501
Arg Val Gly Leu Ala Gln
                165
```

<210> 54
<211> 166
<212> PRT
<213> Brassica napus

<400> 54

```
Ala Leu Asp Ser Asp Asp Val Glu Phe Val Lys Leu Leu Leu Thr Glu
  1               5                  10                  15

Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
             20                  25                  30

Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Thr Leu Asp Met Gly
         35                  40                  45

Asp Val Asn His Arg Asn Ser Arg Gly Tyr Thr Val Leu His Leu Ala
     50                  55                  60

Ala Met Arg Lys Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Lys Gly
 65                  70                  75                  80

Ala Asn Ala Ser Gly Phe Thr Cys Asp Gly Arg Ser Ala Val Asn Ile
                 85                  90                  95

Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ala Ala
            100                 105                 110

Lys Gly Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu
        115                 120                 125

Arg Glu Val Arg Arg Asn Pro Met Val Val Glu Ser Pro Met Cys Ser
    130                 135                 140

Leu Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn
145                 150                 155                 160
```

```
Arg Val Gly Leu Ala Gln
                165


<210> 55
<211> 498
<212> DNA
<213> Brassica napus

<220>
<221> CDS
<222> (2)..(496)
<223> Canola C

<400> 55
g gca ctg gat tct gat gat gtt gag ctt gtg aag ctt ctt ttg acc gag 49
  Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu Leu Thr Glu
    1               5                  10                  15

tca gat atc act cta gat gaa gcc aat ggt ctg cat tac tca gtg gtg   97
Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
            20                  25                  30

tat agt gat ccc aaa gtt gtt gca gag ata ctt gcc ctt ggt tta ggt   145
Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Ala Leu Gly Leu Gly
        35                  40                  45

gat gtc aat cac aga aac tca cgt ggc tac tcg gtt ctt cat ttc gct   193
Asp Val Asn His Arg Asn Ser Arg Gly Tyr Ser Val Leu His Phe Ala
    50                  55                  60

gcc atg cgt aga gag cct tcc atc atc ata tct ctt ctc aag gaa ggc   241
Ala Met Arg Arg Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Glu Gly
65                  70                  75                  80

gcc aat gcg tct agc ttc act ttt gat gga cgc agt gcg gtt aat ata   289
Ala Asn Ala Ser Ser Phe Thr Phe Asp Gly Arg Ser Ala Val Asn Ile
                85                  90                  95

tgt agg aga ctg aca act cca aag gat tat cat aca aag aca tcc aaa   337
Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ser Lys
            100                 105                 110

aag agg gaa gct agt aaa gca agg ctg tgc ata gat ctc ttg gaa aga   385
Lys Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu Arg
        115                 120                 125

gag gtt agg agg aac cct atg ctt gct gat acg cca atg tgt tca ctt   433
Glu Val Arg Arg Asn Pro Met Leu Ala Asp Thr Pro Met Cys Ser Leu
    130                 135                 140

act atg cct gaa gat ctc caa atg aga ctg tta tac ctt gaa aat cga   481
Thr Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggt ctt gct aaa ct                                        498
Val Gly Leu Ala Lys
                165
```

<210> 56
<211> 165
<212> PRT
<213> Brassica napus

<400> 56
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu Leu Thr Glu
1 5 10 15

Ser Asp Ile Thr Leu Asp Glu Ala Asn Gly Leu His Tyr Ser Val Val
20 25 30

Tyr Ser Asp Pro Lys Val Val Ala Glu Ile Leu Ala Leu Gly Leu Gly
35 40 45

Asp Val Asn His Arg Asn Ser Arg Gly Tyr Ser Val Leu His Phe Ala
50 55 60

Ala Met Arg Arg Glu Pro Ser Ile Ile Ile Ser Leu Leu Lys Glu Gly
65 70 75 80

Ala Asn Ala Ser Ser Phe Thr Phe Asp Gly Arg Ser Ala Val Asn Ile
85 90 95

Cys Arg Arg Leu Thr Thr Pro Lys Asp Tyr His Thr Lys Thr Ser Lys
100 105 110

Lys Arg Glu Ala Ser Lys Ala Arg Leu Cys Ile Asp Leu Leu Glu Arg
115 120 125

Glu Val Arg Arg Asn Pro Met Leu Ala Asp Thr Pro Met Cys Ser Leu
130 135 140

Thr Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu Glu Asn Arg
145 150 155 160

Val Gly Leu Ala Lys
165

<210> 57
<211> 498
<212> DNA
<213> Brassica napus

<220>
<221> CDS
<222> (2)..(496)
<223> Canola D

<400> 57
g gca ctg gac tct gat gat gtt gag ctt gtc aag atg ctt ttg aca gaa 49
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu Leu Thr Glu
1 5 10 15

gga cac acg agt cta gac gac gcc tac gct ctt cac tac gct gtt gca 97
Gly His Thr Ser Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
20 25 30

cat tcc gat gtg aag acg gcc tct gat ctc ata gac ctt gag ctt gcg 145

```
His Ser Asp Val Lys Thr Ala Ser Asp Leu Ile Asp Leu Glu Leu Ala
         35                  40                  45

gat gtt gac cat aga aac ctg agg ggg tac acg gcg ctt cac gtt gct   193
Asp Val Asp His Arg Asn Leu Arg Gly Tyr Thr Ala Leu His Val Ala
     50                  55                  60

gcg atg agg aac gag ccg aag ctg atg gtt tat tta ttg act aaa ggt   241
Ala Met Arg Asn Glu Pro Lys Leu Met Val Tyr Leu Leu Thr Lys Gly
 65                  70                  75                  80

gcg aat gcg tcg gag aca acg ttt gac ggt aga acg gct ctt gtg att   289
Ala Asn Ala Ser Glu Thr Thr Phe Asp Gly Arg Thr Ala Leu Val Ile
                 85                  90                  95

gca aaa aga ctc act aaa gct tct gag tat aat gct agt acg gag caa   337
Ala Lys Arg Leu Thr Lys Ala Ser Glu Tyr Asn Ala Ser Thr Glu Gln
            100                 105                 110

ggg aag cct tct ctg aaa gga ggg cta tgc ata gag gta cta gag cat   385
Gly Lys Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val Leu Glu His
        115                 120                 125

gcg cgg aaa cta ggt agg ttg cct aga gat ggt tta cct tct ctt cca   433
Ala Arg Lys Leu Gly Arg Leu Pro Arg Asp Gly Leu Pro Ser Leu Pro
    130                 135                 140

gct act cct gat gaa ctg agg atg agg ttg ctc tac ctt gaa aat cga   481
Ala Thr Pro Asp Glu Leu Arg Met Arg Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

gtt ggc ctg gct caa ct                                           498
Val Gly Leu Ala Gln
                165


<210> 58
<211> 165
<212> PRT
<213> Brassica napus

<400> 58
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu Leu Thr Glu
  1               5                  10                  15

Gly His Thr Ser Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala Val Ala
             20                  25                  30

His Ser Asp Val Lys Thr Ala Ser Asp Leu Ile Asp Leu Glu Leu Ala
         35                  40                  45

Asp Val Asp His Arg Asn Leu Arg Gly Tyr Thr Ala Leu His Val Ala
     50                  55                  60

Ala Met Arg Asn Glu Pro Lys Leu Met Val Tyr Leu Leu Thr Lys Gly
 65                  70                  75                  80

Ala Asn Ala Ser Glu Thr Thr Phe Asp Gly Arg Thr Ala Leu Val Ile
                 85                  90                  95

Ala Lys Arg Leu Thr Lys Ala Ser Glu Tyr Asn Ala Ser Thr Glu Gln
```

```
            100                 105                 110

Gly Lys Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val Leu Glu His
        115                 120                 125

Ala Arg Lys Leu Gly Arg Leu Pro Arg Asp Gly Leu Pro Ser Leu Pro
    130                 135                 140

Ala Thr Pro Asp Glu Leu Arg Met Arg Leu Leu Tyr Leu Glu Asn Arg
145                 150                 155                 160

Val Gly Leu Ala Gln
                165
```

<210> 59
<211> 31
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 3A

<400> 59
tagatgawgc mtaygctcty caytatgctg t 31

<210> 60
<211> 32
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: PCR primer NIM 3B

<400> 60
ggctcyttmc kcatggcagc aayrtgaags ac 32

<210> 61
<211> 148
<212> DNA
<213> Lycopersicon esculentum

<220>
<221> CDS
<222> (4)..(147)
<223> Tomato B

<400> 61

```
tag atg atg cat atg ctc ttc att atg ctg ttg cat att gtg acc cca   48
    Met Met His Met Leu Phe Ile Met Leu Leu His Ile Val Thr Pro
      1               5                  10                  15

agg ttg ttg ctg agg ttc ttg gac tgg gtg ttg cta atg tca acc ttc   96
Arg Leu Leu Leu Arg Phe Leu Asp Trp Val Leu Leu Met Ser Thr Phe
                20                  25                  30
```

```
gga atg cac gtg gtt aca ctg tcc ttc acg ttg ctg cca tgc gga aag   144
Gly Met His Val Val Thr Leu Ser Phe Thr Leu Leu Pro Cys Gly Lys
            35                  40                  45

agc c                                                             148
Ser
```

<210> 62
<211> 48
<212> PRT
<213> Lycopersicon esculentum

<400> 62

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Met Met His Met Leu Phe Ile Met Leu Leu His Ile Val Thr Pro Arg
  1               5                  10                  15

Leu Leu Leu Arg Phe Leu Asp Trp Val Leu Leu Met Ser Thr Phe Gly
             20                  25                  30

Met His Val Val Thr Leu Ser Phe Thr Leu Leu Pro Cys Gly Lys Ser
         35                  40                  45
```

<210> 63
<211> 2296
<212> DNA
<213> Beta vulgaris

<220>
<221> CDS
<222> (113)..(1927)
<223> full-length Sugarbeet cDNA sequence

<400> 63

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cacacacaca cccgacgccg tatgcgtatc cattctctct cctcaacctc cctttgactt 60

cctcttactc caccatcttc aatgtcgtcg atttccaatc tctaacattc ac atg aca 118
                                                          Met Thr
                                                            1

acc acc tcc aca aca atg gtg atc gat tct cgc acc gct ttc tcc gat   166
Thr Thr Ser Thr Thr Met Val Ile Asp Ser Arg Thr Ala Phe Ser Asp
          5                  10                  15

tcc aac gac atc agc aat ggc agt agc atc tgc tgc gtc gcc gca aca   214
Ser Asn Asp Ile Ser Asn Gly Ser Ser Ile Cys Cys Val Ala Ala Thr
     20                  25                  30

aca act aca aca aca acc gcc gca gaa aac tct ctc tcc ttt act ccc   262
Thr Thr Thr Thr Thr Thr Ala Ala Glu Asn Ser Leu Ser Phe Thr Pro
 35                  40                  45                  50

gac gcc gcc gct ctt ctc cgc ctc tct gaa aac ctc gac tcg ctt ttc   310
Asp Ala Ala Ala Leu Leu Arg Leu Ser Glu Asn Leu Asp Ser Leu Phe
                 55                  60                  65

caa ccc tcg ctt tct ctc tcc gac tcc gac tct ttc gcc gac gct aaa   358
Gln Pro Ser Leu Ser Leu Ser Asp Ser Asp Ser Phe Ala Asp Ala Lys
             70                  75                  80
```

```
atc gtc gtt tcc ggt gat tcg cgt gaa gtc gcc gtt cat cgg tgt gtt   406
Ile Val Val Ser Gly Asp Ser Arg Glu Val Ala Val His Arg Cys Val
         85                  90                  95

ctc tcg tct cgg agc tcg ttc ttt cgg tcc gct ttt gct tcg aaa cga   454
Leu Ser Ser Arg Ser Ser Phe Phe Arg Ser Ala Phe Ala Ser Lys Arg
    100                 105                 110

gag aag gag aag gag agg gat aaa gag aga gtg gtg aag ctt gag ctt   502
Glu Lys Glu Lys Glu Arg Asp Lys Glu Arg Val Val Lys Leu Glu Leu
115                 120                 125                 130

aag gat tta gct ggt gat ttt gag gtt gga ttt gat tcg gtt gtt gcg   550
Lys Asp Leu Ala Gly Asp Phe Glu Val Gly Phe Asp Ser Val Val Ala
                135                 140                 145

gtt tta ggt tat ttg tat agt ggc aaa gtt agg aat ttg cct aga gga   598
Val Leu Gly Tyr Leu Tyr Ser Gly Lys Val Arg Asn Leu Pro Arg Gly
            150                 155                 160

att tgt gtt tgt gtt gat gag gat tgc tct cat gaa gct tgt cgt cct   646
Ile Cys Val Cys Val Asp Glu Asp Cys Ser His Glu Ala Cys Arg Pro
        165                 170                 175

gct gtt gat ttt gtt gtt gag gtt ctc tat ttg tct cac aaa ttc gag   694
Ala Val Asp Phe Val Val Glu Val Leu Tyr Leu Ser His Lys Phe Glu
    180                 185                 190

att gtc gaa ttg gtt tcg ctt tat cag agg cac cta ctg gat att ctt   742
Ile Val Glu Leu Val Ser Leu Tyr Gln Arg His Leu Leu Asp Ile Leu
195                 200                 205                 210

gac aag att gca cca gat gac gtt cta gta gtg tta tct gtc gct gag   790
Asp Lys Ile Ala Pro Asp Asp Val Leu Val Val Leu Ser Val Ala Glu
                215                 220                 225

atg tgt gga aat gcg tgt gac gga ttg ctg gca agg tgt att gac aag   838
Met Cys Gly Asn Ala Cys Asp Gly Leu Leu Ala Arg Cys Ile Asp Lys
            230                 235                 240

att gtg agg tcc gat att gac gta acc acc att gat aaa tcc ttg ccg   886
Ile Val Arg Ser Asp Ile Asp Val Thr Thr Ile Asp Lys Ser Leu Pro
        245                 250                 255

cag aat gtt gtg aaa cag ata atc gac acg cga aag gaa ctt ggg ttt   934
Gln Asn Val Val Lys Gln Ile Ile Asp Thr Arg Lys Glu Leu Gly Phe
    260                 265                 270

act gaa cct ggg cgt gtt gag ttt cct gat aag cat gtg aag aga ata   982
Thr Glu Pro Gly Arg Val Glu Phe Pro Asp Lys His Val Lys Arg Ile
275                 280                 285                 290

cac aga gct ttg gaa tcc gat gat gta gag tta gtc aga atg ctt tta   1030
His Arg Ala Leu Glu Ser Asp Asp Val Glu Leu Val Arg Met Leu Leu
                295                 300                 305

aaa gag cgc cat aca act cta gat gat gca tat gcc ctt cac tat gct   1078
Lys Glu Arg His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His Tyr Ala
            310                 315                 320
```

```
gtg gca cat tgt gat gcc aag acc acc acg gag ctt ctt gag ctt ggg   1126
Val Ala His Cys Asp Ala Lys Thr Thr Thr Glu Leu Leu Glu Leu Gly
        325                 330                 335

ctt gca gat gtt aat ctt aga aat cta agg ggt cac act gtg cta cat   1174
Leu Ala Asp Val Asn Leu Arg Asn Leu Arg Gly His Thr Val Leu His
    340                 345                 350

gtg gca gcc atg aga aaa gag cct aag ata att gta tcc ttg tta acc   1222
Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu Leu Thr
355                 360                 365                 370

aag gga gcc cat ccg tct gat ata aca tca gat gat aaa aaa gca ctg   1270
Lys Gly Ala His Pro Ser Asp Ile Thr Ser Asp Asp Lys Lys Ala Leu
                375                 380                 385

cag ata gca aag aga cta aca aaa gct gtg gac ttc tat aaa act aca   1318
Gln Ile Ala Lys Arg Leu Thr Lys Ala Val Asp Phe Tyr Lys Thr Thr
            390                 395                 400

gaa caa gga aaa gat gca cca aag gat cgg ttg tgc att gaa ata ctg   1366
Glu Gln Gly Lys Asp Ala Pro Lys Asp Arg Leu Cys Ile Glu Ile Leu
        405                 410                 415

gag caa gct gaa aga aga gaa cca ttg cta gga gaa ggt tct gtt tct   1414
Glu Gln Ala Glu Arg Arg Glu Pro Leu Leu Gly Glu Gly Ser Val Ser
    420                 425                 430

ctt gca aag gca gga gat gat ctg cgt atg aag cta tta tat ctt gaa   1462
Leu Ala Lys Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr Leu Glu
435                 440                 445                 450

aat aga gtt gca ctt gct cgg ttg ctc ttt cca atg gaa gcg aaa gtg   1510
Asn Arg Val Ala Leu Ala Arg Leu Leu Phe Pro Met Glu Ala Lys Val
                455                 460                 465

gct atg gat att gct caa gtg gac gga act tct gaa ttc aca ttg tca   1558
Ala Met Asp Ile Ala Gln Val Asp Gly Thr Ser Glu Phe Thr Leu Ser
            470                 475                 480

aag aat ata gct gat gca cga aga aat gcg gtg gac ttg aat gag gct   1606
Lys Asn Ile Ala Asp Ala Arg Arg Asn Ala Val Asp Leu Asn Glu Ala
        485                 490                 495

ccc ttt ata ttg aaa gag gag cat ttg cag agg atg aaa gca ctg tct   1654
Pro Phe Ile Leu Lys Glu Glu His Leu Gln Arg Met Lys Ala Leu Ser
    500                 505                 510

aaa act gtt gag ctt ggc aag cgt ttc ttt cca cgc tgc tcc gat gtt   1702
Lys Thr Val Glu Leu Gly Lys Arg Phe Phe Pro Arg Cys Ser Asp Val
515                 520                 525                 530

ctt aat aag att atg gac gcc gaa gat cta tca cag ctt gca ttt tta   1750
Leu Asn Lys Ile Met Asp Ala Glu Asp Leu Ser Gln Leu Ala Phe Leu
                535                 540                 545

gga aaa gat act cca gag gaa cgg caa agg aag aga aaa cga tac ctt   1798
Gly Lys Asp Thr Pro Glu Glu Arg Gln Arg Lys Arg Lys Arg Tyr Leu
            550                 555                 560

gaa ctg caa gac gct tta act aag gct ttt aca gag gac aaa gaa gag   1846
```

```
Glu Leu Gln Asp Ala Leu Thr Lys Ala Phe Thr Glu Asp Lys Glu Glu
        565                 570                 575

ttt gac cgt tct aca tta tca tca tcg tcg tcg tcg act cca atg ggg   1894
Phe Asp Arg Ser Thr Leu Ser Ser Ser Ser Ser Ser Thr Pro Met Gly
    580                 585                 590

agg cca tat ggt aag acc aat ttc aag agg taa ctccttagca gctcaaagtt 1947
Arg Pro Tyr Gly Lys Thr Asn Phe Lys Arg
595                 600                 605

gcatacgacg tcacttgtat aatattcatg tatatgtatg aaaatttctt tttgttctcc 2007

ccttctattg atggccacgg tttcgatctt tttggtctgt attataattt ttgaccgatt 2067

acttgataga attgtattct atacatcttt ataagctcat agtaacacca gatttaggta 2127

ctatccgttg gagacacata ctcttgtgtg cgatgatgaa tcaatcatca gattacatta 2187

cacgagccat ttcctgccat attgtaattc atgtatcaag gtacaaataa atagcgtcgt 2247

ggggttgcac ctcttgcatt atcgaaaaaa aaaaaaaaaa aaaaaaaaa             2296
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<210> 64
<211> 604
<212> PRT
<213> Beta vulgaris

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<400> 64
Met Thr Thr Thr Ser Thr Thr Met Val Ile Asp Ser Arg Thr Ala Phe
  1               5                  10                  15
Ser Asp Ser Asn Asp Ile Ser Asn Gly Ser Ser Ile Cys Cys Val Ala
             20                  25                  30
Ala Thr Thr Thr Thr Thr Thr Thr Ala Ala Glu Asn Ser Leu Ser Phe
         35                  40                  45
Thr Pro Asp Ala Ala Ala Leu Leu Arg Leu Ser Glu Asn Leu Asp Ser
     50                  55                  60
Leu Phe Gln Pro Ser Leu Ser Leu Ser Asp Ser Asp Ser Phe Ala Asp
 65                  70                  75                  80
Ala Lys Ile Val Val Ser Gly Asp Ser Arg Glu Val Ala Val His Arg
                 85                  90                  95
Cys Val Leu Ser Ser Arg Ser Ser Phe Phe Arg Ser Ala Phe Ala Ser
            100                 105                 110
Lys Arg Glu Lys Glu Lys Glu Arg Asp Lys Glu Arg Val Val Lys Leu
        115                 120                 125
Glu Leu Lys Asp Leu Ala Gly Asp Phe Glu Val Gly Phe Asp Ser Val
    130                 135                 140
Val Ala Val Leu Gly Tyr Leu Tyr Ser Gly Lys Val Arg Asn Leu Pro
145                 150                 155                 160
Arg Gly Ile Cys Val Cys Val Asp Glu Asp Cys Ser His Glu Ala Cys
                165                 170                 175
Arg Pro Ala Val Asp Phe Val Val Glu Val Leu Tyr Leu Ser His Lys
            180                 185                 190
Phe Glu Ile Val Glu Leu Val Ser Leu Tyr Gln Arg His Leu Leu Asp
        195                 200                 205
Ile Leu Asp Lys Ile Ala Pro Asp Asp Val Leu Val Val Leu Ser Val
    210                 215                 220
Ala Glu Met Cys Gly Asn Ala Cys Asp Gly Leu Leu Ala Arg Cys Ile
225                 230                 235                 240
Asp Lys Ile Val Arg Ser Asp Ile Asp Val Thr Thr Ile Asp Lys Ser
```

```
                245                 250                 255
Leu Pro Gln Asn Val Val Lys Gln Ile Ile Asp Thr Arg Lys Glu Leu
            260                 265                 270
Gly Phe Thr Glu Pro Gly Arg Val Glu Phe Pro Asp Lys His Val Lys
        275                 280                 285
Arg Ile His Arg Ala Leu Glu Ser Asp Asp Val Glu Leu Val Arg Met
    290                 295                 300
Leu Leu Lys Glu Arg His Thr Thr Leu Asp Asp Ala Tyr Ala Leu His
305                 310                 315                 320
Tyr Ala Val Ala His Cys Asp Ala Lys Thr Thr Thr Glu Leu Leu Glu
                325                 330                 335
Leu Gly Leu Ala Asp Val Asn Leu Arg Asn Leu Arg Gly His Thr Val
            340                 345                 350
Leu His Val Ala Ala Met Arg Lys Glu Pro Lys Ile Ile Val Ser Leu
        355                 360                 365
Leu Thr Lys Gly Ala His Pro Ser Asp Ile Thr Ser Asp Asp Lys Lys
    370                 375                 380
Ala Leu Gln Ile Ala Lys Arg Leu Thr Lys Ala Val Asp Phe Tyr Lys
385                 390                 395                 400
Thr Thr Glu Gln Gly Lys Asp Ala Pro Lys Asp Arg Leu Cys Ile Glu
                405                 410                 415
Ile Leu Glu Gln Ala Glu Arg Arg Glu Pro Leu Leu Gly Glu Gly Ser
            420                 425                 430
Val Ser Leu Ala Lys Ala Gly Asp Asp Leu Arg Met Lys Leu Leu Tyr
        435                 440                 445
Leu Glu Asn Arg Val Ala Leu Ala Arg Leu Leu Phe Pro Met Glu Ala
    450                 455                 460
Lys Val Ala Met Asp Ile Ala Gln Val Asp Gly Thr Ser Glu Phe Thr
465                 470                 475                 480
Leu Ser Lys Asn Ile Ala Asp Ala Arg Arg Asn Ala Val Asp Leu Asn
                485                 490                 495
Glu Ala Pro Phe Ile Leu Lys Glu Glu His Leu Gln Arg Met Lys Ala
            500                 505                 510
Leu Ser Lys Thr Val Glu Leu Gly Lys Arg Phe Phe Pro Arg Cys Ser
        515                 520                 525
Asp Val Leu Asn Lys Ile Met Asp Ala Glu Asp Leu Ser Gln Leu Ala
    530                 535                 540
Phe Leu Gly Lys Asp Thr Pro Glu Glu Arg Gln Arg Lys Arg Lys Arg
545                 550                 555                 560
Tyr Leu Glu Leu Gln Asp Ala Leu Thr Lys Ala Phe Thr Glu Asp Lys
                565                 570                 575
Glu Glu Phe Asp Arg Ser Thr Leu Ser Ser Ser Ser Ser Ser Thr Pro
            580                 585                 590
Met Gly Arg Pro Tyr Gly Lys Thr Asn Phe Lys Arg
        595                 600
```

<210> 65
<211> 2844
<212> DNA
<213> Helianthus annuus

<220>
<221> CDS
<222> (737)..(2512)
<223> full-length Sunflower B cDNA sequence

<400> 65
gacgataaaa ccctctctc tttttgctac caagaacctt cctactttct tgcaccaaag 60

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tttctttgca ggttctttga agcttcttta tcatcatacg ttggtttgat attgtttttg 120

atgcatcttt tcacatgggt tttgcttatt gagtgattat ctgttgtggg tatttgatac 180

aaattgaaaa aaagatgatt agatttggta tttagggttt tggttattga agattttatt 240

aattagggtt tgattagggt ttgattaaga ttcttgtatt ggatgggttg atttagatcc 300

agctgtttgt gggtttcaaa tttttgtttt ggtatttgca tatctcattc taatctattc 360

agaggttgag gttctttagg tttgactttg actttgactt ttgggtactt tcttgtacat 420

gtataatgtt tgatttgatc cattatatgt gttttgtaat tgaatcatag caaattttct 480

tgcctgtata tatatgtttt attgaggatt tggttcaagt tttgaccttt ttgggaaaaa 540

aagtcaaaca catattcttg ttcatgtagt tttgcaaatc aatcatttca caaatctttc 600

tttatgttgg gaatccatct caatcataaa aaagtttctt tctttctttg agttcttgtt 660

agctatgaaa gtttatgatt tgtccttttt gtgataaagt caaaccccta atcatcctgg 720

gactttgact aaatcg atg gcg aat tca tcc gaa ccg tca tca tcc ata agc 772
                Met Ala Asn Ser Ser Glu Pro Ser Ser Ser Ile Ser
                  1               5                  10

ttc acc tca tct tca cac ata tct aac ggc gca act agc tac aac ata   820
Phe Thr Ser Ser Ser His Ile Ser Asn Gly Ala Thr Ser Tyr Asn Ile
         15                  20                  25

ccc cca cca tca atc ccc gag cca cgg tcg aat att gaa atc att ggc   868
Pro Pro Pro Ser Ile Pro Glu Pro Arg Ser Asn Ile Glu Ile Ile Gly
     30                  35                  40

tta aat aga ctc agc aca aac cta gag aag ctc gta ttc gat tca ggt   916
Leu Asn Arg Leu Ser Thr Asn Leu Glu Lys Leu Val Phe Asp Ser Gly
 45                  50                  55                  60

tct gaa tct gat tgc aat tac agc gat gct gaa gtt gtt gtt gag ggt   964
Ser Glu Ser Asp Cys Asn Tyr Ser Asp Ala Glu Val Val Val Glu Gly
                 65                  70                  75

att tct gta ggc att cat cgg tgt att tta gcc act aga agt acg ttt   1012
Ile Ser Val Gly Ile His Arg Cys Ile Leu Ala Thr Arg Ser Thr Phe
             80                  85                  90

ttt agc gat ttg ttt aag aag aac aaa ggt tgt gta gag aag gac agt   1060
Phe Ser Asp Leu Phe Lys Lys Asn Lys Gly Cys Val Glu Lys Asp Ser
         95                  100                 105

aag ccg aaa tat aac atg agt gat ttg ttg ccg tat ggg agc gtt ggg   1108
Lys Pro Lys Tyr Asn Met Ser Asp Leu Leu Pro Tyr Gly Ser Val Gly
    110                 115                 120

tat gat gcg ttt ctc gtg ttt tta agc tat gtt tat act ggg aaa ctg   1156
Tyr Asp Ala Phe Leu Val Phe Leu Ser Tyr Val Tyr Thr Gly Lys Leu
125                 130                 135                 140

aaa gcg tct cct ccg gag gtt tca acc tgc gtt gat gat ggg tgt ctt   1204
Lys Ala Ser Pro Pro Glu Val Ser Thr Cys Val Asp Asp Gly Cys Leu
                145                 150                 155
```

```
cat gat gct tgt tgg cct gct att aac ttt gct gtt gag ttg act tat   1252
His Asp Ala Cys Trp Pro Ala Ile Asn Phe Ala Val Glu Leu Thr Tyr
            160                 165                 170

gcg tct tcg gtt ttt caa gtt ccg gaa tta gtt tcg ctt ttt cag cgt   1300
Ala Ser Ser Val Phe Gln Val Pro Glu Leu Val Ser Leu Phe Gln Arg
        175                 180                 185

cgt ctt ctc aac ttt gtg gac aag gct ctt gtt gaa gac gtg atc ccg   1348
Arg Leu Leu Asn Phe Val Asp Lys Ala Leu Val Glu Asp Val Ile Pro
    190                 195                 200

atc ctt gtt gtg gcc ttt cac tgt cag ttg caa aac gtc tta tct cgt   1396
Ile Leu Val Val Ala Phe His Cys Gln Leu Gln Asn Val Leu Ser Arg
205                 210                 215                 220

tgc att gac cga gta gtt agg tca aag ctc gat act att tcc att gaa   1444
Cys Ile Asp Arg Val Val Arg Ser Lys Leu Asp Thr Ile Ser Ile Glu
                225                 230                 235

aaa gag ctt cca ttt gaa gtc acc caa atg atc aaa tcc att gat aac   1492
Lys Glu Leu Pro Phe Glu Val Thr Gln Met Ile Lys Ser Ile Asp Asn
            240                 245                 250

atc atc caa gaa gat gac gaa cat aca gtc gaa tca gaa gtc gtg tta   1540
Ile Ile Gln Glu Asp Asp Glu His Thr Val Glu Ser Glu Val Val Leu
        255                 260                 265

cgt gaa aag aga att aaa agc ata cac aaa gca tta gac tgt gac gat   1588
Arg Glu Lys Arg Ile Lys Ser Ile His Lys Ala Leu Asp Cys Asp Asp
    270                 275                 280

gtt gag ctt gtg aaa atg att tta gac gaa tcc aaa atc acg tta gat   1636
Val Glu Leu Val Lys Met Ile Leu Asp Glu Ser Lys Ile Thr Leu Asp
285                 290                 295                 300

gaa gcc tgc gct ctt cat tat gcg gtc atg tat tgt aat caa gaa gtt   1684
Glu Ala Cys Ala Leu His Tyr Ala Val Met Tyr Cys Asn Gln Glu Val
                305                 310                 315

gct aag gag att ctt aac tta aac cgt gcg gat gtt aat ctt aga aac   1732
Ala Lys Glu Ile Leu Asn Leu Asn Arg Ala Asp Val Asn Leu Arg Asn
            320                 325                 330

tca cga gat tac acc gtg ctt cat gtt gct gcc atg cgt aaa gaa cca   1780
Ser Arg Asp Tyr Thr Val Leu His Val Ala Ala Met Arg Lys Glu Pro
        335                 340                 345

tca ctt att gtt tcg att cta agc aaa ggc gcg tgt gca tcg gat act   1828
Ser Leu Ile Val Ser Ile Leu Ser Lys Gly Ala Cys Ala Ser Asp Thr
    350                 355                 360

act ttt gat gga caa agt gcg gtt agt att tgc agg aga cga aca agg   1876
Thr Phe Asp Gly Gln Ser Ala Val Ser Ile Cys Arg Arg Arg Thr Arg
365                 370                 375                 380

ccc aag gat tat tat gtg aaa acc gaa cac ggg caa gaa aca aat aaa   1924
Pro Lys Asp Tyr Tyr Val Lys Thr Glu His Gly Gln Glu Thr Asn Lys
                385                 390                 395
```

```
gat cgt ata tgc atc gat gtt ttg gag cgg gaa ata aag agg aat ccg   1972
Asp Arg Ile Cys Ile Asp Val Leu Glu Arg Glu Ile Lys Arg Asn Pro
            400                 405                 410

atg ata ggc gat gtt tcc gtg tgt tct tca gca gtg gct gat gat ttg   2020
Met Ile Gly Asp Val Ser Val Cys Ser Ser Ala Val Ala Asp Asp Leu
        415                 420                 425

cat atg aat tta ctc tac tta gaa aac cga gtg gca ttt gct cga ctg   2068
His Met Asn Leu Leu Tyr Leu Glu Asn Arg Val Ala Phe Ala Arg Leu
    430                 435                 440

tta ttt ccg tca gaa gcg aaa cta gca atg gaa att gcg cat gcc caa   2116
Leu Phe Pro Ser Glu Ala Lys Leu Ala Met Glu Ile Ala His Ala Gln
445                 450                 455                 460

acg act gca cag tat ccg ggt cta ttg gca tcg aaa ggg tca aat ggt   2164
Thr Thr Ala Gln Tyr Pro Gly Leu Leu Ala Ser Lys Gly Ser Asn Gly
                465                 470                 475

aac tta agg gag atg gat ttg aac gag aca ccg ttg gtg cag aac aaa   2212
Asn Leu Arg Glu Met Asp Leu Asn Glu Thr Pro Leu Val Gln Asn Lys
            480                 485                 490

aga ttg ctt tca aga atg gaa gcc ctt tcc cgg aca gtg gaa atg ggt   2260
Arg Leu Leu Ser Arg Met Glu Ala Leu Ser Arg Thr Val Glu Met Gly
        495                 500                 505

agg cga tat ttc cct cat tgt tca gag gtt ctg gat aag ttc atg gag   2308
Arg Arg Tyr Phe Pro His Cys Ser Glu Val Leu Asp Lys Phe Met Glu
    510                 515                 520

gac gat cta cag gat ctt ttt atc ctc gag aag ggt acc gaa gaa gaa   2356
Asp Asp Leu Gln Asp Leu Phe Ile Leu Glu Lys Gly Thr Glu Glu Glu
525                 530                 535                 540

caa gaa atc aaa agg acg cga ttt atg gag ctt aaa gaa gat gtc caa   2404
Gln Glu Ile Lys Arg Thr Arg Phe Met Glu Leu Lys Glu Asp Val Gln
                545                 550                 555

aga gcc ttt acc aag gac aag gcc gag ctt cat cgc ggt ttg tcc tca   2452
Arg Ala Phe Thr Lys Asp Lys Ala Glu Leu His Arg Gly Leu Ser Ser
            560                 565                 570

tca atg tac acc ccc aca gtg aga aac ggg tca aag agt aaa gcc cgc   2500
Ser Met Tyr Thr Pro Thr Val Arg Asn Gly Ser Lys Ser Lys Ala Arg
        575                 580                 585

aaa tac tca tga aacccccgtg tttctttgat gatcttttaa cacgctttta       2552
Lys Tyr Ser
    590

cgtgcctaat attagaggca aaacatatgt atgaagaaat aatggtggtg catgatgatg 2612

tttagggctc aggtttaggg tttatatgta ctaaattttg tgatttgacg ctaaaaatgc 2672

tatgttgttt ttttttttt ttggataata tggtgtgaaa gctaacgcct tttactagta 2732

gcatgttaat gtttgtgttt gaatcatagt tttttatgca tgtttgtttt acttgcacaa 2792

caactaataa atataatttt tcataataaa aaaaaaaaaa aaaaaaaaaa aa          2844
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<210> 66
<211> 591
<212> PRT
<213> Helianthus annuus

<400> 66

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Met Ala Asn Ser Ser Glu Pro Ser Ser Ser Ile Ser Phe Thr Ser Ser
 1               5                  10                  15
Ser His Ile Ser Asn Gly Ala Thr Ser Tyr Asn Ile Pro Pro Pro Ser
            20                  25                  30
Ile Pro Glu Pro Arg Ser Asn Ile Glu Ile Ile Gly Leu Asn Arg Leu
        35                  40                  45
Ser Thr Asn Leu Glu Lys Leu Val Phe Asp Ser Gly Ser Glu Ser Asp
    50                  55                  60
Cys Asn Tyr Ser Asp Ala Glu Val Val Val Glu Gly Ile Ser Val Gly
65                  70                  75                  80
Ile His Arg Cys Ile Leu Ala Thr Arg Ser Thr Phe Phe Ser Asp Leu
                85                  90                  95
Phe Lys Lys Asn Lys Gly Cys Val Glu Lys Asp Ser Lys Pro Lys Tyr
            100                 105                 110
Asn Met Ser Asp Leu Leu Pro Tyr Gly Ser Val Gly Tyr Asp Ala Phe
        115                 120                 125
Leu Val Phe Leu Ser Tyr Val Tyr Thr Gly Lys Leu Lys Ala Ser Pro
    130                 135                 140
Pro Glu Val Ser Thr Cys Val Asp Asp Gly Cys Leu His Asp Ala Cys
145                 150                 155                 160
Trp Pro Ala Ile Asn Phe Ala Val Glu Leu Thr Tyr Ala Ser Ser Val
                165                 170                 175
Phe Gln Val Pro Glu Leu Val Ser Leu Phe Gln Arg Arg Leu Leu Asn
            180                 185                 190
Phe Val Asp Lys Ala Leu Val Glu Asp Val Ile Pro Ile Leu Val Val
        195                 200                 205
Ala Phe His Cys Gln Leu Gln Asn Val Leu Ser Arg Cys Ile Asp Arg
    210                 215                 220
Val Val Arg Ser Lys Leu Asp Thr Ile Ser Ile Glu Lys Glu Leu Pro
225                 230                 235                 240
Phe Glu Val Thr Gln Met Ile Lys Ser Ile Asp Asn Ile Ile Gln Glu
                245                 250                 255
Asp Asp Glu His Thr Val Glu Ser Glu Val Val Leu Arg Glu Lys Arg
            260                 265                 270
Ile Lys Ser Ile His Lys Ala Leu Asp Cys Asp Asp Val Glu Leu Val
        275                 280                 285
Lys Met Ile Leu Asp Glu Ser Lys Ile Thr Leu Asp Glu Ala Cys Ala
    290                 295                 300
Leu His Tyr Ala Val Met Tyr Cys Asn Gln Glu Val Ala Lys Glu Ile
305                 310                 315                 320
Leu Asn Leu Asn Arg Ala Asp Val Asn Leu Arg Asn Ser Arg Asp Tyr
                325                 330                 335
Thr Val Leu His Val Ala Ala Met Arg Lys Glu Pro Ser Leu Ile Val
            340                 345                 350
Ser Ile Leu Ser Lys Gly Ala Cys Ala Ser Asp Thr Thr Phe Asp Gly
        355                 360                 365
Gln Ser Ala Val Ser Ile Cys Arg Arg Arg Thr Arg Pro Lys Asp Tyr
    370                 375                 380
Tyr Val Lys Thr Glu His Gly Gln Glu Thr Asn Lys Asp Arg Ile Cys
385                 390                 395                 400
Ile Asp Val Leu Glu Arg Glu Ile Lys Arg Asn Pro Met Ile Gly Asp
                405                 410                 415
Val Ser Val Cys Ser Ser Ala Val Ala Asp Asp Leu His Met Asn Leu
```

```
            420                 425                 430
Leu Tyr Leu Glu Asn Arg Val Ala Phe Ala Arg Leu Leu Phe Pro Ser
        435                 440                 445
Glu Ala Lys Leu Ala Met Glu Ile Ala His Ala Gln Thr Thr Ala Gln
    450                 455                 460
Tyr Pro Gly Leu Leu Ala Ser Lys Gly Ser Asn Gly Asn Leu Arg Glu
465                 470                 475                 480
Met Asp Leu Asn Glu Thr Pro Leu Val Gln Asn Lys Arg Leu Leu Ser
                485                 490                 495
Arg Met Glu Ala Leu Ser Arg Thr Val Glu Met Gly Arg Arg Tyr Phe
            500                 505                 510
Pro His Cys Ser Glu Val Leu Asp Lys Phe Met Glu Asp Asp Leu Gln
        515                 520                 525
Asp Leu Phe Ile Leu Glu Lys Gly Thr Glu Glu Glu Gln Glu Ile Lys
    530                 535                 540
Arg Thr Arg Phe Met Glu Leu Lys Glu Asp Val Gln Arg Ala Phe Thr
545                 550                 555                 560
Lys Asp Lys Ala Glu Leu His Arg Gly Leu Ser Ser Ser Met Tyr Thr
                565                 570                 575
Pro Thr Val Arg Asn Gly Ser Lys Ser Lys Ala Arg Lys Tyr Ser
            580                 585                 590
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<210> 67
<211> 1477
<212> DNA
<213> Arabidopsis thaliana

<220>
<221> CDS
<222> (1)..(804)
<223> AtNMLc2 cDNA sequence

<220>
<221> misc_feature
<222> (1)..(1477)
<223> n = a, t, c or g

<400> 67

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atg agc aat ctt gaa gaa tct ttg aga tct cta tcg ttg gat ttc ctg   48
Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
  1               5                  10                  15

aac cta cta atc aac ggt caa gct ttc tcc gac gtg act ttc agc gtt   96
Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
             20                  25                  30

gaa ggt cgt tta gtc cac gct cac cgt tgt atc ctc gcc gca cgg agg   144
Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Arg
         35                  40                  45

ctt ttc ttc cgc aaa ttc ttt tgt ggg aca gac tca cca caa cct gtc   192
Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
     50                  55                  60

aca ggt ata gac ccg acc caa cat ggg tcc gta ccc gct agc cca aca   240
Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
 65                  70                  75                  80

aga ggc tcc acg gcc cca gct gga att ata cca gtg aac tca gtc ggt   288
```

```
Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val Asn Ser Val Gly
                 85                  90                  95

tat gag gtt ttt ctg ttg cta ctt cag ttt ctt tat agc gga caa gtc   336
Tyr Glu Val Phe Leu Leu Leu Leu Gln Phe Leu Tyr Ser Gly Gln Val
            100                 105                 110

tcc atc gtg ccg cag aaa cac gag cct aga cct aat tgt ggc gag aga   384
Ser Ile Val Pro Gln Lys His Glu Pro Arg Pro Asn Cys Gly Glu Arg
        115                 120                 125

gga tgt tgg cac act cat tgc tca gcc gcc gtt gat ctt gct ctt gat   432
Gly Cys Trp His Thr His Cys Ser Ala Ala Val Asp Leu Ala Leu Asp
    130                 135                 140

act ctc gcc gcc tct cgt tac ttc ggc gtc gag cag ctc gca ttg ctc   480
Thr Leu Ala Ala Ser Arg Tyr Phe Gly Val Glu Gln Leu Ala Leu Leu
145                 150                 155                 160

acc cag aaa caa ttg gca agc atg gtg gag aaa gcc tct atc gaa gat   528
Thr Gln Lys Gln Leu Ala Ser Met Val Glu Lys Ala Ser Ile Glu Asp
                165                 170                 175

gtg atg aaa gtt tta ata gca tca aga aag caa gac atg cat caa tta   576
Val Met Lys Val Leu Ile Ala Ser Arg Lys Gln Asp Met His Gln Leu
            180                 185                 190

tgg acc acc tgc tct cac tta gtt gct aaa tca ggt ctc cca cca gag   624
Trp Thr Thr Cys Ser His Leu Val Ala Lys Ser Gly Leu Pro Pro Glu
        195                 200                 205

att ctt gcc aag cat ctc cct att gac gtc gtc acc aaa ata gaa gag   672
Ile Leu Ala Lys His Leu Pro Ile Asp Val Val Thr Lys Ile Glu Glu
    210                 215                 220

ctt cgt ctt aaa tct tct ata gct cgc cgt tct cta atg cct cac aac   720
Leu Arg Leu Lys Ser Ser Ile Ala Arg Arg Ser Leu Met Pro His Asn
225                 230                 235                 240

cac cac cat gat ctc agc ggn gnt caa nac cta aag ntc aaa gtt aga   768
His His His Asp Leu Ser Xaa Xaa Gln Xaa Leu Lys Xaa Lys Val Arg
                245                 250                 255

agg ttg agc cga ctt gga ttc ttc aac gng aac tag taaagctgat        814
Arg Leu Ser Arg Leu Gly Phe Phe Asn Xaa Asn
            260                 265

ggtaatggan aaggactcca ttcttgatga agtcgtaagc attgcattac cgcttgttaa 874

aagctgtaga agagaagttg tgaagncttt ngcttgaagc ttggaagctg ccgatgtgaa 934

ttatccggcg ggtccggcaa ggnaaancac ctttgcactt cgcgggntga gatggtctct 994

ccagacatgg tggctgttct gttagcccnc catgcttgat cctaatgtga ggacagttgg 1054

tggaatcacg cctcttgata tccttagaac attaacttcg gatttcttgt tcaaggggca 1114

gttcctggat tgactcacat tgaaccgaat aaacttaggc tttgcctcga gcttgttcaa 1174

tccgctgcaa tggtgatatc tcgagaagaa ggaaacaata gcaacaacca aaacaatgat 1234
```

aacaataccg ggatttaccc tcatatgaat gaggagcaca atagtggaag cagtggaggg 1294

agcaataaca atttggattc aagattggtt tatctcaatc ttggagcagg tacgggtcag 1354

atgggtccag gtcgagatca aggggatgac cataacagtc agagggaagg tatgagtcgg 1414

catcatcatc atcatcaaga cccatctaca atgtatcatc accatcatca acatcacttc 1474

tag 1477

<210> 68
<211> 267
<212> PRT
<213> Arabidopsis thaliana

<400> 68

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Met Ser Asn Leu Glu Glu Ser Leu Arg Ser Leu Ser Leu Asp Phe Leu
  1               5                  10                  15
Asn Leu Leu Ile Asn Gly Gln Ala Phe Ser Asp Val Thr Phe Ser Val
             20                  25                  30
Glu Gly Arg Leu Val His Ala His Arg Cys Ile Leu Ala Ala Arg Arg
         35                  40                  45
Leu Phe Phe Arg Lys Phe Phe Cys Gly Thr Asp Ser Pro Gln Pro Val
     50                  55                  60
Thr Gly Ile Asp Pro Thr Gln His Gly Ser Val Pro Ala Ser Pro Thr
 65                  70                  75                  80
Arg Gly Ser Thr Ala Pro Ala Gly Ile Ile Pro Val Asn Ser Val Gly
                 85                  90                  95
Tyr Glu Val Phe Leu Leu Leu Leu Gln Phe Leu Tyr Ser Gly Gln Val
            100                 105                 110
Ser Ile Val Pro Gln Lys His Glu Pro Arg Pro Asn Cys Gly Glu Arg
        115                 120                 125
Gly Cys Trp His Thr His Cys Ser Ala Ala Val Asp Leu Ala Leu Asp
    130                 135                 140
Thr Leu Ala Ala Ser Arg Tyr Phe Gly Val Glu Gln Leu Ala Leu Leu
145                 150                 155                 160
Thr Gln Lys Gln Leu Ala Ser Met Val Glu Lys Ala Ser Ile Glu Asp
                165                 170                 175
Val Met Lys Val Leu Ile Ala Ser Arg Lys Gln Asp Met His Gln Leu
            180                 185                 190
Trp Thr Thr Cys Ser His Leu Val Ala Lys Ser Gly Leu Pro Pro Glu
        195                 200                 205
Ile Leu Ala Lys His Leu Pro Ile Asp Val Val Thr Lys Ile Glu Glu
    210                 215                 220
Leu Arg Leu Lys Ser Ser Ile Ala Arg Arg Ser Leu Met Pro His Asn
225                 230                 235                 240
His His His Asp Leu Ser Xaa Xaa Gln Xaa Leu Lys Xaa Lys Val Arg
                245                 250                 255
Arg Leu Ser Arg Leu Gly Phe Phe Asn Xaa Asn
            260                 265
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<210> 69
<211> 1725
<212> DNA
<213> Arabidopsis thaliana

<220>
<221> CDS

<222> (1)..(1725)
<223> AtNMLc4-1 cDNA sequence

<400> 69

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atg gct gca act gca ata gag cca tct tca tct ata agt ttc aca tct   48
Met Ala Ala Thr Ala Ile Glu Pro Ser Ser Ser Ile Ser Phe Thr Ser
  1               5                  10                  15

tct cac tta tca aac cct tct cct gtt gtt act act tat cac tca gct   96
Ser His Leu Ser Asn Pro Ser Pro Val Val Thr Thr Tyr His Ser Ala
             20                  25                  30

gcc aat ctt gaa gag ctc agc tct aac ttg gag cag ctt ctc act aat   144
Ala Asn Leu Glu Glu Leu Ser Ser Asn Leu Glu Gln Leu Leu Thr Asn
         35                  40                  45

cca gat tgc gat tac act gac gca gag atc atc att gaa gaa gaa gct   192
Pro Asp Cys Asp Tyr Thr Asp Ala Glu Ile Ile Ile Glu Glu Glu Ala
     50                  55                  60

aac cct gtg agt gtt cat aga tgt gtt tta gct gct agg agc aag ttt   240
Asn Pro Val Ser Val His Arg Cys Val Leu Ala Ala Arg Ser Lys Phe
 65                  70                  75                  80

ttt ctt gat ctg ttt aag aaa gat aaa gat agt agt gag aag aaa cct   288
Phe Leu Asp Leu Phe Lys Lys Asp Lys Asp Ser Ser Glu Lys Lys Pro
                 85                  90                  95

aag tat caa atg aaa gat tta tta cca tat gga aat gtg gga cgt gag   336
Lys Tyr Gln Met Lys Asp Leu Leu Pro Tyr Gly Asn Val Gly Arg Glu
            100                 105                 110

gca ttt ctg cat ttc ttg agc tat atc tac act ggg agg tta aag cct   384
Ala Phe Leu His Phe Leu Ser Tyr Ile Tyr Thr Gly Arg Leu Lys Pro
        115                 120                 125

ttt cct atc gag gtt tca act tgt gtt gat tca gtt tgt gct cat gat   432
Phe Pro Ile Glu Val Ser Thr Cys Val Asp Ser Val Cys Ala His Asp
    130                 135                 140

tct tgt aaa ccg gcc att gat ttt gct gtt gag ttg atg tat gct tca   480
Ser Cys Lys Pro Ala Ile Asp Phe Ala Val Glu Leu Met Tyr Ala Ser
145                 150                 155                 160

ttt gtg ttc caa atc ccg gat ctt gtt tcg tca ttt cag cgg aag ctt   528
Phe Val Phe Gln Ile Pro Asp Leu Val Ser Ser Phe Gln Arg Lys Leu
                165                 170                 175

cgt aac tat gtt gag aag tca cta gta gag aat gtt ctt cct atc ctc   576
Arg Asn Tyr Val Glu Lys Ser Leu Val Glu Asn Val Leu Pro Ile Leu
            180                 185                 190

tta gtt gcg ttt cat tgt gat ttg aca cag ctt ctt gat caa tgc att   624
Leu Val Ala Phe His Cys Asp Leu Thr Gln Leu Leu Asp Gln Cys Ile
        195                 200                 205

gag aga gtg gcg aga tca gac tta gac aga ttc tgt atc gaa aag gag   672
Glu Arg Val Ala Arg Ser Asp Leu Asp Arg Phe Cys Ile Glu Lys Glu
    210                 215                 220

ctt cct tta gaa gta ttg gaa aaa atc aaa cag ctt cga gtt aag tcg   720
```

```
Leu Pro Leu Glu Val Leu Glu Lys Ile Lys Gln Leu Arg Val Lys Ser
225                 230                 235                 240

gtg aac ata ccc gag gtg gag gat aaa tcg ata gag aga aca ggg aaa   768
Val Asn Ile Pro Glu Val Glu Asp Lys Ser Ile Glu Arg Thr Gly Lys
                245                 250                 255

gta ctc aag gca ttg gat tca gat gat gta gaa ctc gtg aag ctt ctt   816
Val Leu Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
            260                 265                 270

ttg act gag tca gat ata act cta gac caa gcc aat ggt cta cat tat   864
Leu Thr Glu Ser Asp Ile Thr Leu Asp Gln Ala Asn Gly Leu His Tyr
        275                 280                 285

gca gtg gca tac agt gat ccg aaa gtt gtg aca cag gtt ctt gat cta   912
Ala Val Ala Tyr Ser Asp Pro Lys Val Val Thr Gln Val Leu Asp Leu
    290                 295                 300

gat atg gct gat gtt aat ttc aga aat tcc agg ggg tat acg gtt ctt   960
Asp Met Ala Asp Val Asn Phe Arg Asn Ser Arg Gly Tyr Thr Val Leu
305                 310                 315                 320

cat att gct gct atg cgt aga gag cca aca att atc ata cca ctt att  1008
His Ile Ala Ala Met Arg Arg Glu Pro Thr Ile Ile Ile Pro Leu Ile
                325                 330                 335

caa aaa gga gct aat gct tca gat ttc acg ttt gat gga cgc agt gcg  1056
Gln Lys Gly Ala Asn Ala Ser Asp Phe Thr Phe Asp Gly Arg Ser Ala
            340                 345                 350

gta aat ata tgt agg aga ctc act agg ccg aaa gat tat cat acc aaa  1104
Val Asn Ile Cys Arg Arg Leu Thr Arg Pro Lys Asp Tyr His Thr Lys
        355                 360                 365

acc tca agg aaa gaa cct agt aaa tac cgc tta tgc atc gat atc ttg  1152
Thr Ser Arg Lys Glu Pro Ser Lys Tyr Arg Leu Cys Ile Asp Ile Leu
    370                 375                 380

gaa agg gaa att aga agg aat cca ttg gtt agt ggg gat aca ccc act  1200
Glu Arg Glu Ile Arg Arg Asn Pro Leu Val Ser Gly Asp Thr Pro Thr
385                 390                 395                 400

tgt tcc cat tcg atg ccc gag gat ctc caa atg agg ttg tta tac tta  1248
Cys Ser His Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu
                405                 410                 415

gaa aag cga gtg gga ctt gct cag ttg ttc ttc cca gca gaa gcc aat  1296
Glu Lys Arg Val Gly Leu Ala Gln Leu Phe Phe Pro Ala Glu Ala Asn
            420                 425                 430

gtg gct atg gac gtt gct aat gtt gaa ggg aca agc gag tgc aca ggt  1344
Val Ala Met Asp Val Ala Asn Val Glu Gly Thr Ser Glu Cys Thr Gly
        435                 440                 445

ctt cta act cca cct cca tca aat gat aca act gaa aac ttg ggt aaa  1392
Leu Leu Thr Pro Pro Pro Ser Asn Asp Thr Thr Glu Asn Leu Gly Lys
    450                 455                 460

gtc gat tta aat gaa acg cct tat gtg caa acg aaa aga atg ctt aca  1440
Val Asp Leu Asn Glu Thr Pro Tyr Val Gln Thr Lys Arg Met Leu Thr
```

```
465                 470                 475                 480

cgt atg aaa gcc ctc atg aaa aca gtt gag aca ggt cgg aga tac ttc   1488
Arg Met Lys Ala Leu Met Lys Thr Val Glu Thr Gly Arg Arg Tyr Phe
                485                 490                 495

cca tct tgt tat gag gtt ctg gat aag tac atg gat cag tat atg gac   1536
Pro Ser Cys Tyr Glu Val Leu Asp Lys Tyr Met Asp Gln Tyr Met Asp
            500                 505                 510

gaa gaa atc cct gat atg tcg tat ccc gag aaa ggc act gtg aaa gag   1584
Glu Glu Ile Pro Asp Met Ser Tyr Pro Glu Lys Gly Thr Val Lys Glu
        515                 520                 525

aga aga cag aag agg atg aga tat aac gag ctg aag aac gac gtt aaa   1632
Arg Arg Gln Lys Arg Met Arg Tyr Asn Glu Leu Lys Asn Asp Val Lys
    530                 535                 540

aaa gca tat agc aaa gac aaa gtc gcg cgg tct tgt ctt tct tct tca   1680
Lys Ala Tyr Ser Lys Asp Lys Val Ala Arg Ser Cys Leu Ser Ser Ser
545                 550                 555                 560

tca cca gct tct tct ctt aga gaa gcc tta gag aat cca aca tga       1725
Ser Pro Ala Ser Ser Leu Arg Glu Ala Leu Glu Asn Pro Thr
                565                 570                 575
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<210> 70
<211> 574
<212> PRT
<213> Arabidopsis thaliana

<400> 70

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Met Ala Ala Thr Ala Ile Glu Pro Ser Ser Ser Ile Ser Phe Thr Ser
  1               5                  10                  15
Ser His Leu Ser Asn Pro Ser Pro Val Val Thr Thr Tyr His Ser Ala
            20                  25                  30
Ala Asn Leu Glu Glu Leu Ser Ser Asn Leu Glu Gln Leu Leu Thr Asn
        35                  40                  45
Pro Asp Cys Asp Tyr Thr Asp Ala Glu Ile Ile Ile Glu Glu Glu Ala
    50                  55                  60
Asn Pro Val Ser Val His Arg Cys Val Leu Ala Ala Arg Ser Lys Phe
65                  70                  75                  80
Phe Leu Asp Leu Phe Lys Lys Asp Lys Asp Ser Ser Glu Lys Lys Pro
                85                  90                  95
Lys Tyr Gln Met Lys Asp Leu Leu Pro Tyr Gly Asn Val Gly Arg Glu
            100                 105                 110
Ala Phe Leu His Phe Leu Ser Tyr Ile Tyr Thr Gly Arg Leu Lys Pro
        115                 120                 125
Phe Pro Ile Glu Val Ser Thr Cys Val Asp Ser Val Cys Ala His Asp
    130                 135                 140
Ser Cys Lys Pro Ala Ile Asp Phe Ala Val Glu Leu Met Tyr Ala Ser
145                 150                 155                 160
Phe Val Phe Gln Ile Pro Asp Leu Val Ser Ser Phe Gln Arg Lys Leu
                165                 170                 175
Arg Asn Tyr Val Glu Lys Ser Leu Val Glu Asn Val Leu Pro Ile Leu
            180                 185                 190
Leu Val Ala Phe His Cys Asp Leu Thr Gln Leu Leu Asp Gln Cys Ile
        195                 200                 205
Glu Arg Val Ala Arg Ser Asp Leu Asp Arg Phe Cys Ile Glu Lys Glu
    210                 215                 220
```

```
Leu Pro Leu Glu Val Leu Glu Lys Ile Lys Gln Leu Arg Val Lys Ser
225                 230                 235                 240
Val Asn Ile Pro Glu Val Glu Asp Lys Ser Ile Glu Arg Thr Gly Lys
                245                 250                 255
Val Leu Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
            260                 265                 270
Leu Thr Glu Ser Asp Ile Thr Leu Asp Gln Ala Asn Gly Leu His Tyr
        275                 280                 285
Ala Val Ala Tyr Ser Asp Pro Lys Val Val Thr Gln Val Leu Asp Leu
    290                 295                 300
Asp Met Ala Asp Val Asn Phe Arg Asn Ser Arg Gly Tyr Thr Val Leu
305                 310                 315                 320
His Ile Ala Ala Met Arg Arg Glu Pro Thr Ile Ile Ile Pro Leu Ile
                325                 330                 335
Gln Lys Gly Ala Asn Ala Ser Asp Phe Thr Phe Asp Gly Arg Ser Ala
            340                 345                 350
Val Asn Ile Cys Arg Arg Leu Thr Arg Pro Lys Asp Tyr His Thr Lys
        355                 360                 365
Thr Ser Arg Lys Glu Pro Ser Lys Tyr Arg Leu Cys Ile Asp Ile Leu
    370                 375                 380
Glu Arg Glu Ile Arg Arg Asn Pro Leu Val Ser Gly Asp Thr Pro Thr
385                 390                 395                 400
Cys Ser His Ser Met Pro Glu Asp Leu Gln Met Arg Leu Leu Tyr Leu
                405                 410                 415
Glu Lys Arg Val Gly Leu Ala Gln Leu Phe Phe Pro Ala Glu Ala Asn
            420                 425                 430
Val Ala Met Asp Val Ala Asn Val Glu Gly Thr Ser Glu Cys Thr Gly
        435                 440                 445
Leu Leu Thr Pro Pro Pro Ser Asn Asp Thr Thr Glu Asn Leu Gly Lys
    450                 455                 460
Val Asp Leu Asn Glu Thr Pro Tyr Val Gln Thr Lys Arg Met Leu Thr
465                 470                 475                 480
Arg Met Lys Ala Leu Met Lys Thr Val Glu Thr Gly Arg Arg Tyr Phe
                485                 490                 495
Pro Ser Cys Tyr Glu Val Leu Asp Lys Tyr Met Asp Gln Tyr Met Asp
            500                 505                 510
Glu Glu Ile Pro Asp Met Ser Tyr Pro Glu Lys Gly Thr Val Lys Glu
        515                 520                 525
Arg Arg Gln Lys Arg Met Arg Tyr Asn Glu Leu Lys Asn Asp Val Lys
    530                 535                 540
Lys Ala Tyr Ser Lys Asp Lys Val Ala Arg Ser Cys Leu Ser Ser Ser
545                 550                 555                 560
Ser Pro Ala Ser Ser Leu Arg Glu Ala Leu Glu Asn Pro Thr
                565                 570
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<210> 71
<211> 1818
<212> DNA
<213> Arabidopsis thaliana

<220>
<221> CDS
<222> (13)..(1818)
<223> AtNMLc4-2 cDNA sequence

<400> 71

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gccgatctcg tg atg atg gcc acc acc acc acc acc acc gct aga ttc 51
              Met Met Ala Thr Thr Thr Thr Thr Thr Thr Ala Arg Phe
                1               5                  10
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tct gat tca tac gag ttc agc aac aca agc ggc aat agc ttc ttc gcc   99
Ser Asp Ser Tyr Glu Phe Ser Asn Thr Ser Gly Asn Ser Phe Phe Ala
     15                  20                  25

gcc gag tca tct ctt gat tat ccg acg gaa ttt ctc acg cca ccg gag   147
Ala Glu Ser Ser Leu Asp Tyr Pro Thr Glu Phe Leu Thr Pro Pro Glu
 30                  35                  40                  45

gta tca gct ctt aaa ctt ctg tct aac tgc ctc gag tct gtt ttc gac   195
Val Ser Ala Leu Lys Leu Leu Ser Asn Cys Leu Glu Ser Val Phe Asp
                 50                  55                  60

tcg ccg gag acg ttc tac agc gat gct aag cta gtt ctc gcc ggc ggc   243
Ser Pro Glu Thr Phe Tyr Ser Asp Ala Lys Leu Val Leu Ala Gly Gly
             65                  70                  75

cgg gaa gtt tct ttt cac cgt tgt att ctt tcc gcg aga att cct gtc   291
Arg Glu Val Ser Phe His Arg Cys Ile Leu Ser Ala Arg Ile Pro Val
         80                  85                  90

ttc aaa agc gct tta gcc acc gtg aag gaa caa aaa tcc tcc acc acc   339
Phe Lys Ser Ala Leu Ala Thr Val Lys Glu Gln Lys Ser Ser Thr Thr
     95                  100                 105

gtg aag ctc cag ctg aaa gag atc gcc aga gat tac gaa gtc ggc ttt   387
Val Lys Leu Gln Leu Lys Glu Ile Ala Arg Asp Tyr Glu Val Gly Phe
110                 115                 120                 125

gac tcg gtt gtg gcg gtt ttg gcg tat gtt tac agc ggc aga gtg agg   435
Asp Ser Val Val Ala Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg
                130                 135                 140

tcc ccg ccg aag gga gct tct gct tgc gta gac gac gat tgt tgc cac   483
Ser Pro Pro Lys Gly Ala Ser Ala Cys Val Asp Asp Asp Cys Cys His
            145                 150                 155

gtg gct tgc cgg tca aag gtg gat ttc atg gtg gag gtt ctt tat ctg   531
Val Ala Cys Arg Ser Lys Val Asp Phe Met Val Glu Val Leu Tyr Leu
        160                 165                 170

tct ttc gtt ttc cag att caa gaa tta gtt act ctg tat gag agg cag   579
Ser Phe Val Phe Gln Ile Gln Glu Leu Val Thr Leu Tyr Glu Arg Gln
    175                 180                 185

ttc ttg gaa att gta gac aaa gtt gta gtc gaa gac atc ttg gtt ata   627
Phe Leu Glu Ile Val Asp Lys Val Val Val Glu Asp Ile Leu Val Ile
190                 195                 200                 205

ttc aag ctt gat act cta tgt ggt aca aca tac aag aag ctt ttg gat   675
Phe Lys Leu Asp Thr Leu Cys Gly Thr Thr Tyr Lys Lys Leu Leu Asp
                210                 215                 220

aga tgc ata gaa att atc gtg aag tct gat ata gaa cta gtt agt ctt   723
Arg Cys Ile Glu Ile Ile Val Lys Ser Asp Ile Glu Leu Val Ser Leu
            225                 230                 235

gag aag tct tta cct caa cac att ttc aag caa atc ata gac atc cgc   771
Glu Lys Ser Leu Pro Gln His Ile Phe Lys Gln Ile Ile Asp Ile Arg
        240                 245                 250
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gaa gcg ctc tgt cta gag cca cct aaa cta gaa agg cat gtc aag aac   819
Glu Ala Leu Cys Leu Glu Pro Pro Lys Leu Glu Arg His Val Lys Asn
    255                 260                 265

ata tac aag gcg cta gac tca gat gat gtt gag ctt gtc aag atg ctt   867
Ile Tyr Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu
270                 275                 280                 285

ttg cta gaa gga cac acc aat ctc gat gag gcg tat gct ctt cat ttt   915
Leu Leu Glu Gly His Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe
                290                 295                 300

gct atc gct cac tgc gct gtg aag acc gcg tat gat ctc ctc gag ctt   963
Ala Ile Ala His Cys Ala Val Lys Thr Ala Tyr Asp Leu Leu Glu Leu
            305                 310                 315

gag ctt gcg gat gtt aac ctt aga aat ccg agg gga tac act gtg ctt   1011
Glu Leu Ala Asp Val Asn Leu Arg Asn Pro Arg Gly Tyr Thr Val Leu
        320                 325                 330

cat gtt gct gcg atg cgg aag gag ccg aag ttg ata ata tct ttg tta   1059
His Val Ala Ala Met Arg Lys Glu Pro Lys Leu Ile Ile Ser Leu Leu
    335                 340                 345

atg aaa ggg gca aat att tta gac aca aca ttg gat ggt aga acc gct   1107
Met Lys Gly Ala Asn Ile Leu Asp Thr Thr Leu Asp Gly Arg Thr Ala
350                 355                 360                 365

tta gtg att gta aaa cga ctc act aaa gcg gat gac tac aaa act agt   1155
Leu Val Ile Val Lys Arg Leu Thr Lys Ala Asp Asp Tyr Lys Thr Ser
                370                 375                 380

acg gag gac ggt acg cct tct ctg aaa ggc gga tta tgc ata gag gta   1203
Thr Glu Asp Gly Thr Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val
            385                 390                 395

ctt gag cat gaa caa aaa cta gaa tat ttg tcg cct ata gag gct tca   1251
Leu Glu His Glu Gln Lys Leu Glu Tyr Leu Ser Pro Ile Glu Ala Ser
        400                 405                 410

ctt tct ctt cca gta act cca gag gag ttg agg atg agg ttg ctc tat   1299
Leu Ser Leu Pro Val Thr Pro Glu Glu Leu Arg Met Arg Leu Leu Tyr
    415                 420                 425

tat gaa aac cga gtt gca ctt gct cga ctt ctc ttt cca gtg gaa act   1347
Tyr Glu Asn Arg Val Ala Leu Ala Arg Leu Leu Phe Pro Val Glu Thr
430                 435                 440                 445

gaa act gta cag ggt att gcc aaa ttg gag gaa aca tgc gag ttt aca   1395
Glu Thr Val Gln Gly Ile Ala Lys Leu Glu Glu Thr Cys Glu Phe Thr
                450                 455                 460

gct tct agt ctc gag cct gat cat cac att ggt gaa aag cgg aca tca   1443
Ala Ser Ser Leu Glu Pro Asp His His Ile Gly Glu Lys Arg Thr Ser
            465                 470                 475

cta gac cta aat atg gcg ccg ttc caa atc cat gag aag cat ttg agt   1491
Leu Asp Leu Asn Met Ala Pro Phe Gln Ile His Glu Lys His Leu Ser
        480                 485                 490

aga cta aga gca ctt tgt aaa acc gtg gaa ctg ggg aaa cgc tac ttc   1539
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Arg Leu Arg Ala Leu Cys Lys Thr Val Glu Leu Gly Lys Arg Tyr Phe
    495                 500                 505

aaa cga tgt tcg ctt gat cac ttt atg gat act gag gac ttg aat cat   1587
Lys Arg Cys Ser Leu Asp His Phe Met Asp Thr Glu Asp Leu Asn His
510                 515                 520                 525

ctt gct agc gta gaa gaa gat act cct gag aaa cgg cta caa aag aag   1635
Leu Ala Ser Val Glu Glu Asp Thr Pro Glu Lys Arg Leu Gln Lys Lys
                530                 535                 540

caa agg tac atg gaa cta caa gag act ctg atg aag acc ttt agt gag   1683
Gln Arg Tyr Met Glu Leu Gln Glu Thr Leu Met Lys Thr Phe Ser Glu
            545                 550                 555

gac aag gag gaa tgt gga aag tct tcc aca ccg aaa cca acc tct gcg   1731
Asp Lys Glu Glu Cys Gly Lys Ser Ser Thr Pro Lys Pro Thr Ser Ala
        560                 565                 570

gtg agg tct aat aga aaa ctc tct cac cgg cgc cta aaa gtg gac aaa   1779
Val Arg Ser Asn Arg Lys Leu Ser His Arg Arg Leu Lys Val Asp Lys
    575                 580                 585

cgg gat ttt ttg aaa cga cct tac ggg aac ggg gat taa               1818
Arg Asp Phe Leu Lys Arg Pro Tyr Gly Asn Gly Asp
590                 595                 600
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            20                  25                  30
Ser Leu Asp Tyr Pro Thr Glu Phe Leu Thr Pro Pro Glu Val Ser Ala
        35                  40                  45
Leu Lys Leu Leu Ser Asn Cys Leu Glu Ser Val Phe Asp Ser Pro Glu
    50                  55                  60
Thr Phe Tyr Ser Asp Ala Lys Leu Val Leu Ala Gly Gly Arg Glu Val
65                  70                  75                  80
Ser Phe His Arg Cys Ile Leu Ser Ala Arg Ile Pro Val Phe Lys Ser
                85                  90                  95
Ala Leu Ala Thr Val Lys Glu Gln Lys Ser Ser Thr Thr Val Lys Leu
            100                 105                 110
Gln Leu Lys Glu Ile Ala Arg Asp Tyr Glu Val Gly Phe Asp Ser Val
        115                 120                 125
Val Ala Val Leu Ala Tyr Val Tyr Ser Gly Arg Val Arg Ser Pro Pro
    130                 135                 140
Lys Gly Ala Ser Ala Cys Val Asp Asp Asp Cys Cys His Val Ala Cys
145                 150                 155                 160
Arg Ser Lys Val Asp Phe Met Val Glu Val Leu Tyr Leu Ser Phe Val
                165                 170                 175
Phe Gln Ile Gln Glu Leu Val Thr Leu Tyr Glu Arg Gln Phe Leu Glu
            180                 185                 190
Ile Val Asp Lys Val Val Val Glu Asp Ile Leu Val Ile Phe Lys Leu
        195                 200                 205
Asp Thr Leu Cys Gly Thr Thr Tyr Lys Lys Leu Leu Asp Arg Cys Ile
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    210                 215                 220
Glu Ile Ile Val Lys Ser Asp Ile Glu Leu Val Ser Leu Glu Lys Ser
225                 230                 235                 240
Leu Pro Gln His Ile Phe Lys Gln Ile Ile Asp Ile Arg Glu Ala Leu
                245                 250                 255
Cys Leu Glu Pro Pro Lys Leu Glu Arg His Val Lys Asn Ile Tyr Lys
            260                 265                 270
Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Met Leu Leu Leu Glu
        275                 280                 285
Gly His Thr Asn Leu Asp Glu Ala Tyr Ala Leu His Phe Ala Ile Ala
    290                 295                 300
His Cys Ala Val Lys Thr Ala Tyr Asp Leu Leu Glu Leu Glu Leu Ala
305                 310                 315                 320
Asp Val Asn Leu Arg Asn Pro Arg Gly Tyr Thr Val Leu His Val Ala
                325                 330                 335
Ala Met Arg Lys Glu Pro Lys Leu Ile Ile Ser Leu Leu Met Lys Gly
            340                 345                 350
Ala Asn Ile Leu Asp Thr Thr Leu Asp Gly Arg Thr Ala Leu Val Ile
        355                 360                 365
Val Lys Arg Leu Thr Lys Ala Asp Asp Tyr Lys Thr Ser Thr Glu Asp
    370                 375                 380
Gly Thr Pro Ser Leu Lys Gly Gly Leu Cys Ile Glu Val Leu Glu His
385                 390                 395                 400
Glu Gln Lys Leu Glu Tyr Leu Ser Pro Ile Glu Ala Ser Leu Ser Leu
                405                 410                 415
Pro Val Thr Pro Glu Glu Leu Arg Met Arg Leu Leu Tyr Tyr Glu Asn
            420                 425                 430
Arg Val Ala Leu Ala Arg Leu Leu Phe Pro Val Glu Thr Glu Thr Val
        435                 440                 445
Gln Gly Ile Ala Lys Leu Glu Glu Thr Cys Glu Phe Thr Ala Ser Ser
    450                 455                 460
Leu Glu Pro Asp His His Ile Gly Glu Lys Arg Thr Ser Leu Asp Leu
465                 470                 475                 480
Asn Met Ala Pro Phe Gln Ile His Glu Lys His Leu Ser Arg Leu Arg
                485                 490                 495
Ala Leu Cys Lys Thr Val Glu Leu Gly Lys Arg Tyr Phe Lys Arg Cys
            500                 505                 510
Ser Leu Asp His Phe Met Asp Thr Glu Asp Leu Asn His Leu Ala Ser
        515                 520                 525
Val Glu Glu Asp Thr Pro Glu Lys Arg Leu Gln Lys Lys Gln Arg Tyr
    530                 535                 540
Met Glu Leu Gln Glu Thr Leu Met Lys Thr Phe Ser Glu Asp Lys Glu
545                 550                 555                 560
Glu Cys Gly Lys Ser Ser Thr Pro Lys Pro Thr Ser Ala Val Arg Ser
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Leu Lys Arg Pro Tyr Gly Asn Gly Asp
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ttttttagcc gaagtgaatg ttattccaat tgggtaagct gtgatcaagc agttgaagtt 180

ttttgttgca aaatttgcca gttatcttga ctttttgtga agttggtaaa tttttcattt 240

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gttttgttga ttatagatgg ggtggaattg ttaatttctt ctaaagtttt aaagggttga 360

tttggtttta cctgaaatag ggagaatatg acttgtagtt ttggaatttg cttcttttct 420

tggtctgcat agttgaatgt tattagaaaa cttatggaaa gttttggtca aacttttgtc 480

ctttgagaag aatttcttgt attggtgatt ggttatggtc ttggagaggt tctttttttt 540

tttgcataga gcctgtgcgg agaatattat acatggttaa aaacattaga ttttctggac 600

tttgactatc ttagatgtag ataaattttg tatatgtttt tagaccatta gaattgggaa 660

atg gct tgt tct gct gaa cca tca tca tct ata agc ttt act tca tct 708
Met Ala Cys Ser Ala Glu Pro Ser Ser Ser Ile Ser Phe Thr Ser Ser
1 5 10 15

tcc att aca tcg aat ggg tcg att ggc gtt ggc caa aac act cat gct 756
Ser Ile Thr Ser Asn Gly Ser Ile Gly Val Gly Gln Asn Thr His Ala
20 25 30

tat ggc ggc tct gag aca ggg agt agt tat gaa atc atc agc ttg agt 804
Tyr Gly Gly Ser Glu Thr Gly Ser Ser Tyr Glu Ile Ile Ser Leu Ser
35 40 45

aaa ctc agt aac aat tta gag caa ctc ttg tca gat tcc agc tct gat 852
Lys Leu Ser Asn Asn Leu Glu Gln Leu Leu Ser Asp Ser Ser Ser Asp
50 55 60

ttt act gat gct gag att gtt gtt gag ggt gtt tca ctt ggt gtt cac 900
Phe Thr Asp Ala Glu Ile Val Val Glu Gly Val Ser Leu Gly Val His
65 70 75 80

cgt tgt ata tta gct gcc agg agt aaa ttt ttt cag gat ctt ttt agg 948
Arg Cys Ile Leu Ala Ala Arg Ser Lys Phe Phe Gln Asp Leu Phe Arg
85 90 95

aaa gag aag gga agt tgt gga aag gaa ggt aaa cca aga tat tct atg 996
Lys Glu Lys Gly Ser Cys Gly Lys Glu Gly Lys Pro Arg Tyr Ser Met
100 105 110

acc gat att ttg cct tat ggt aag gtt gga tat gag gct ttc gtt acc 1044
Thr Asp Ile Leu Pro Tyr Gly Lys Val Gly Tyr Glu Ala Phe Val Thr
115 120 125

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ttc cta agc tat ttg tac tca gga aaa ttg aag cat ttc cct ccg gag   1092
Phe Leu Ser Tyr Leu Tyr Ser Gly Lys Leu Lys His Phe Pro Pro Glu
    130                 135                 140

gta tca aca tgt atg gac act ata tgt gct cat gac tct tgc aga cca   1140
Val Ser Thr Cys Met Asp Thr Ile Cys Ala His Asp Ser Cys Arg Pro
145                 150                 155                 160

gca att aat ttt agt gtg gag ttg atg tat gcc tct tcc atg ttt cag   1188
Ala Ile Asn Phe Ser Val Glu Leu Met Tyr Ala Ser Ser Met Phe Gln
                165                 170                 175

gtt cca gag cta gta tca ctt ttc ctg aga cgc ctt atc aat ttt gtt   1236
Val Pro Glu Leu Val Ser Leu Phe Leu Arg Arg Leu Ile Asn Phe Val
            180                 185                 190

ggg aag gct ctt gtg gaa gat gtt atc cca ata ctt aga gtt gct ttt   1284
Gly Lys Ala Leu Val Glu Asp Val Ile Pro Ile Leu Arg Val Ala Phe
        195                 200                 205

cat tgc caa ttg agc gag ctt ctc act cat tcc gtt gat aga gta gca   1332
His Cys Gln Leu Ser Glu Leu Leu Thr His Ser Val Asp Arg Val Ala
    210                 215                 220

cga tca gat ctt gaa atc aca tgc att gag aaa gag gtt ccc ttt gaa   1380
Arg Ser Asp Leu Glu Ile Thr Cys Ile Glu Lys Glu Val Pro Phe Glu
225                 230                 235                 240

gtt gca gag aat att aaa tta ttg tgg ccg aaa tgt cag gtt gat gaa   1428
Val Ala Glu Asn Ile Lys Leu Leu Trp Pro Lys Cys Gln Val Asp Glu
                245                 250                 255

agt aag gtt cta cct gtg gat ccc ttg cat gaa aag aga aaa aat agg   1476
Ser Lys Val Leu Pro Val Asp Pro Leu His Glu Lys Arg Lys Asn Arg
            260                 265                 270

ata tac aag gca ttg gat tcg gat gat gtt gaa ctt gtc aag ctt cta   1524
Ile Tyr Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
        275                 280                 285

ctg agt gag tct aac ata agc tta gat gaa gcc tac gct ctt cat tat   1572
Leu Ser Glu Ser Asn Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr
    290                 295                 300

gct gtg gca tat tgt gat ccc aag gtt gtg act gag gtt ctt gga ctg   1620
Ala Val Ala Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu
305                 310                 315                 320

ggt gtt gcg gat gtc aac cta cgt aat act cgt ggt tac act gtg ctt   1668
Gly Val Ala Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu
                325                 330                 335

cac att gct tcc atg cgt aag gag cca gca gta att gta tcg ctt ttg   1716
His Ile Ala Ser Met Arg Lys Glu Pro Ala Val Ile Val Ser Leu Leu
            340                 345                 350

act aag gga gct cgt gca tca gag act aca ttg gat ggg cag agt gct   1764
Thr Lys Gly Ala Arg Ala Ser Glu Thr Thr Leu Asp Gly Gln Ser Ala
        355                 360                 365

gtt agtatctgta ggaggctgac taggcctaag gagtaccatg caaaaacaga        1817
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Val

acaaggccag gaagcaaaca aagatcgggt atgtattgat gttttggaga gagagatgcg 1877

tcgcaaccca atggctggag atgcattgtt ttcttcccca atgttggccg atgatctgca 1937

catgaaactg cactacctgg aaaatagagt ggcatttgca cggttactgt tccctcttga 1997

agccagacta gccatgcaaa ttgcaaatgc tgagactgca gctgaagtag cagtccgttt 2057

ggcatctaaa agtacatctg ggaacttgag ggaggttgat ttgaatgaga cacccataaa 2117

gcagaaagaa agacttcttt caaggatgca agccctctcg aagacagttg aacttggcaa 2177

gcgctatttt ccacattgct ctcaagttct ggacaagttt atggaggatg acttacccga 2237

cttaattttc cttgagatgg gccctccaga ggagcaaaag atcaagagga agcgatttaa 2297

ggagctcaaa gatgacgttc ancgggcatt taacaaagac aaagctgaac ttcattgctc 2357

ccgcttgtcc tcatcatcat gttcctcttc ttttaaagat ggngcaagtg tcaaacttag 2417

gaaactatga gtaaataggg ttttgtccta tagtttctct nccatctcag ttttgaatgt 2477

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tcgctatcca tttccctatt cagcttgtta aacttccatg tttncagtag aaagaaattt 2597

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Tyr Gly Gly Ser Glu Thr Gly Ser Ser Tyr Glu Ile Ile Ser Leu Ser
        35                  40                  45

Lys Leu Ser Asn Asn Leu Glu Gln Leu Leu Ser Asp Ser Ser Ser Asp
    50                  55                  60

Phe Thr Asp Ala Glu Ile Val Val Glu Gly Val Ser Leu Gly Val His
65                  70                  75                  80

Arg Cys Ile Leu Ala Ala Arg Ser Lys Phe Phe Gln Asp Leu Phe Arg
                85                  90                  95

Lys Glu Lys Gly Ser Cys Gly Lys Glu Gly Lys Pro Arg Tyr Ser Met
            100                 105                 110

Thr Asp Ile Leu Pro Tyr Gly Lys Val Gly Tyr Glu Ala Phe Val Thr
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        115                 120                 125

Phe Leu Ser Tyr Leu Tyr Ser Gly Lys Leu Lys His Phe Pro Pro Glu
    130                 135                 140

Val Ser Thr Cys Met Asp Thr Ile Cys Ala His Asp Ser Cys Arg Pro
145                 150                 155                 160

Ala Ile Asn Phe Ser Val Glu Leu Met Tyr Ala Ser Ser Met Phe Gln
                165                 170                 175

Val Pro Glu Leu Val Ser Leu Phe Leu Arg Arg Leu Ile Asn Phe Val
            180                 185                 190

Gly Lys Ala Leu Val Glu Asp Val Ile Pro Ile Leu Arg Val Ala Phe
        195                 200                 205

His Cys Gln Leu Ser Glu Leu Leu Thr His Ser Val Asp Arg Val Ala
    210                 215                 220

Arg Ser Asp Leu Glu Ile Thr Cys Ile Glu Lys Glu Val Pro Phe Glu
225                 230                 235                 240

Val Ala Glu Asn Ile Lys Leu Leu Trp Pro Lys Cys Gln Val Asp Glu
                245                 250                 255

Ser Lys Val Leu Pro Val Asp Pro Leu His Glu Lys Arg Lys Asn Arg
            260                 265                 270

Ile Tyr Lys Ala Leu Asp Ser Asp Asp Val Glu Leu Val Lys Leu Leu
        275                 280                 285

Leu Ser Glu Ser Asn Ile Ser Leu Asp Glu Ala Tyr Ala Leu His Tyr
    290                 295                 300

Ala Val Ala Tyr Cys Asp Pro Lys Val Val Thr Glu Val Leu Gly Leu
305                 310                 315                 320

Gly Val Ala Asp Val Asn Leu Arg Asn Thr Arg Gly Tyr Thr Val Leu
                325                 330                 335

His Ile Ala Ser Met Arg Lys Glu Pro Ala Val Ile Val Ser Leu Leu
            340                 345                 350

Thr Lys Gly Ala Arg Ala Ser Glu Thr Thr Leu Asp Gly Gln Ser Ala
        355                 360                 365

Val
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